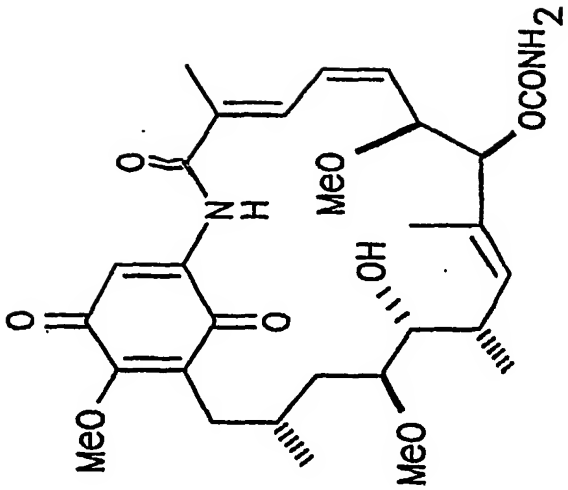
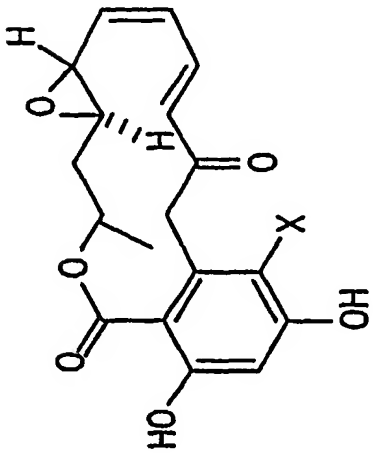
FIG. 1



Geldanamycin (3)



X=Cl Radicicol (1)

X=H Monocillin I (2)

FIG.2

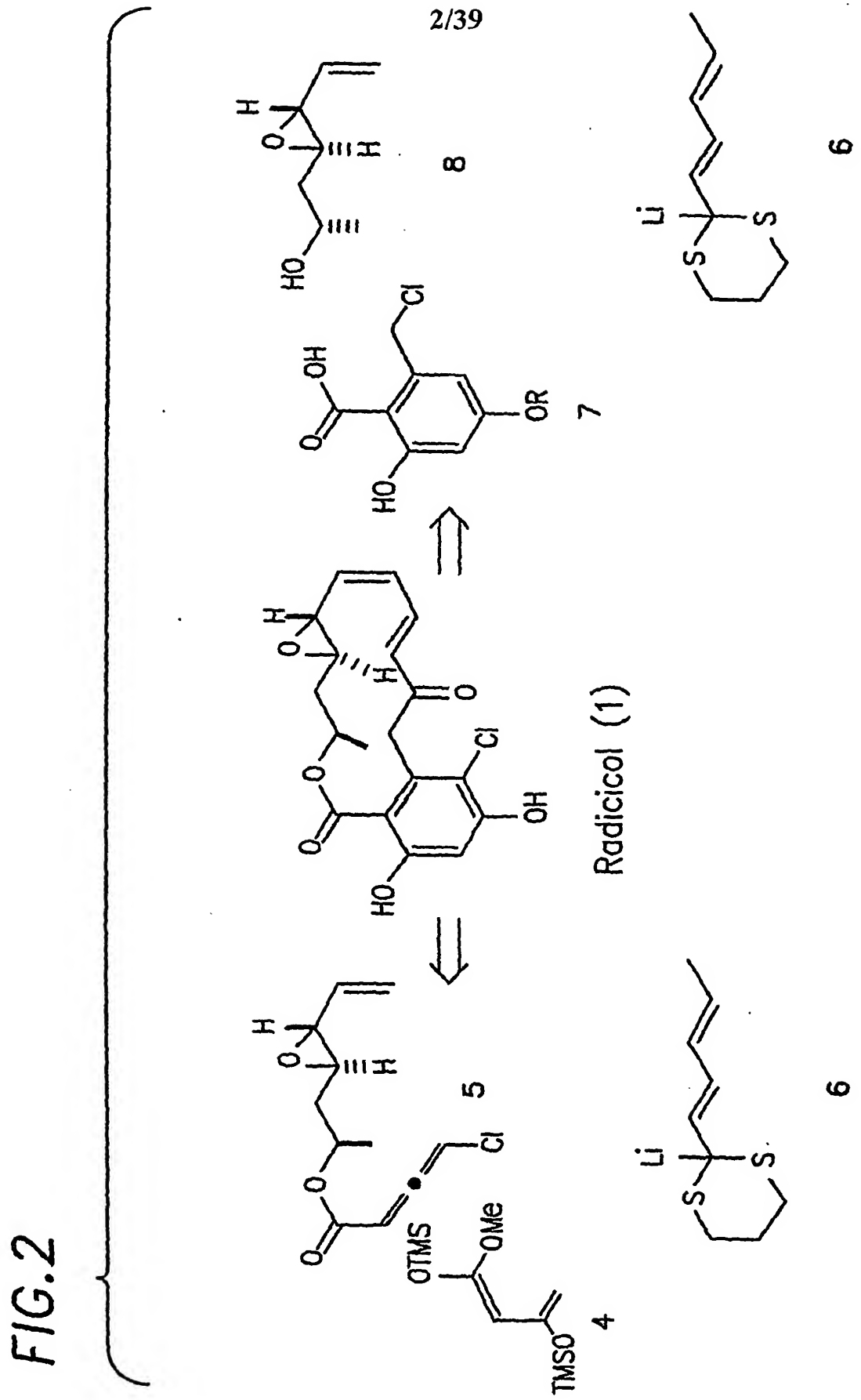
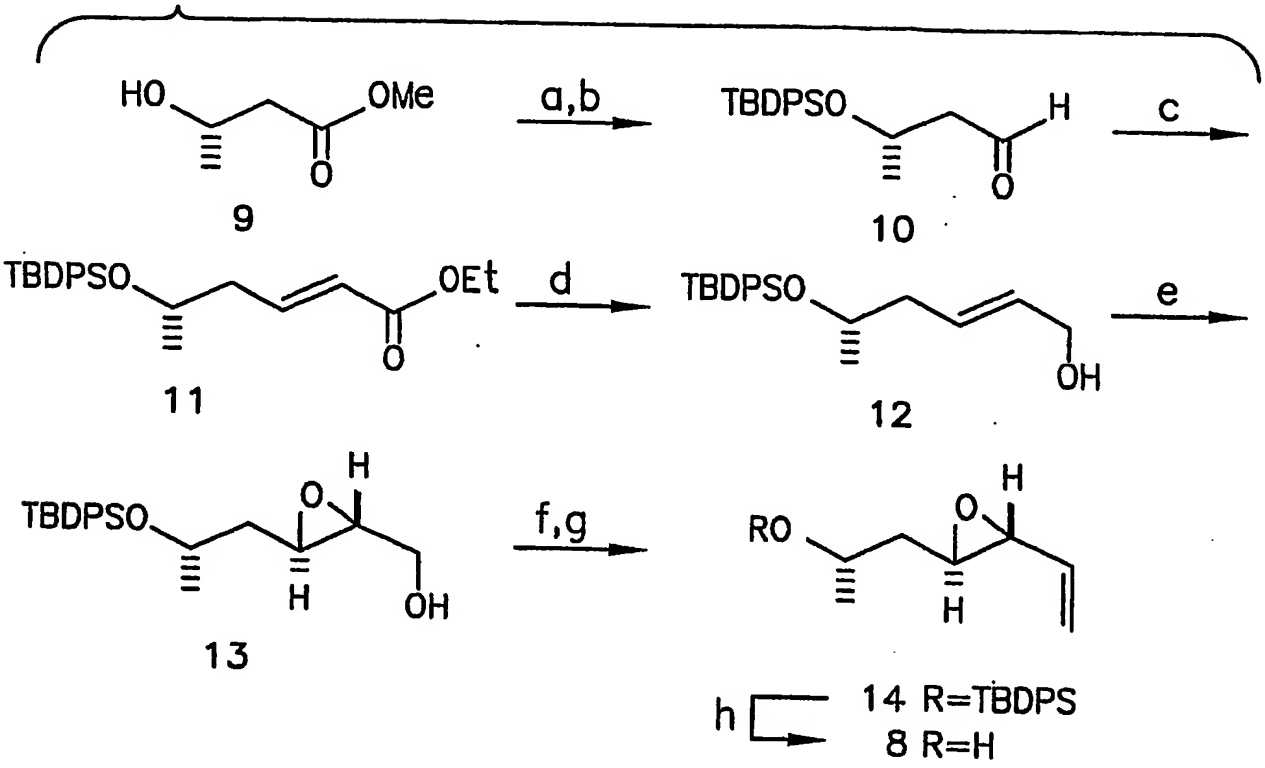
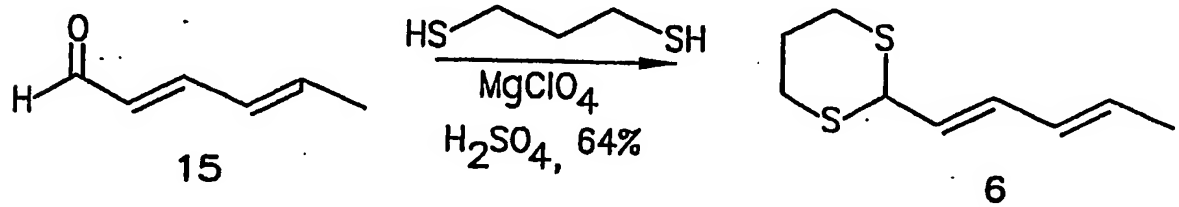


FIG. 3



(a) TBDPSCl, imid.,>95%; (b) DIBAL–H,–78 °C, 92%;
(c) LiCl, DIPEA $(EtO)_2P(O)CH_2CO_2Et$, 95%;
(d) DIBAL–H, –20 °C, 96%; (e) (+)–DET, $Ti(OiPr_4)$, TBHP,90%,>95%ee; (f) SO_3*pyridine, Et_3N, DMSO, 90%;
(g) PH_3PCH_3Br, NaHMDS, 0 °C, 82%; (h) TBAF, 89%.



15 6

FIG. 4

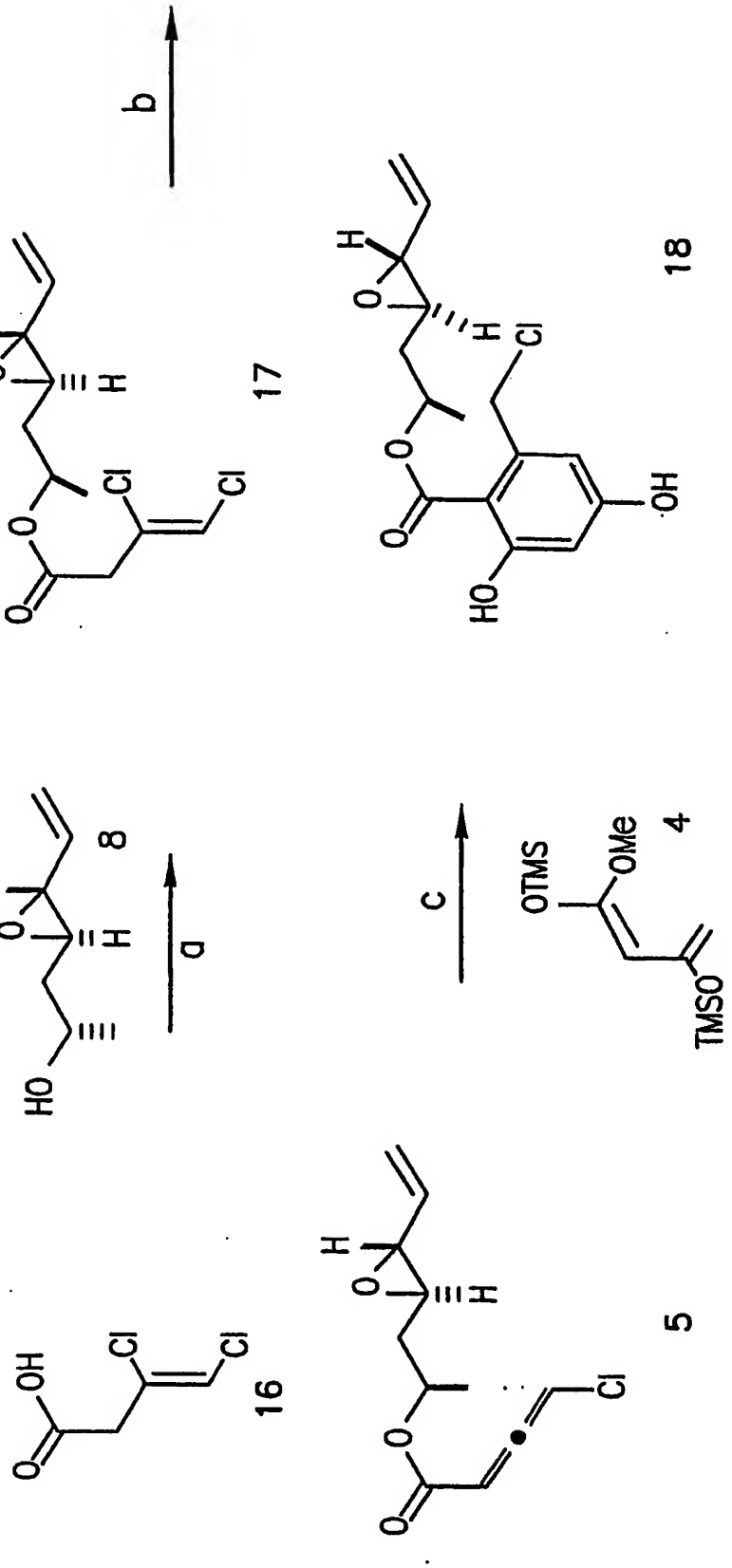


FIG.5

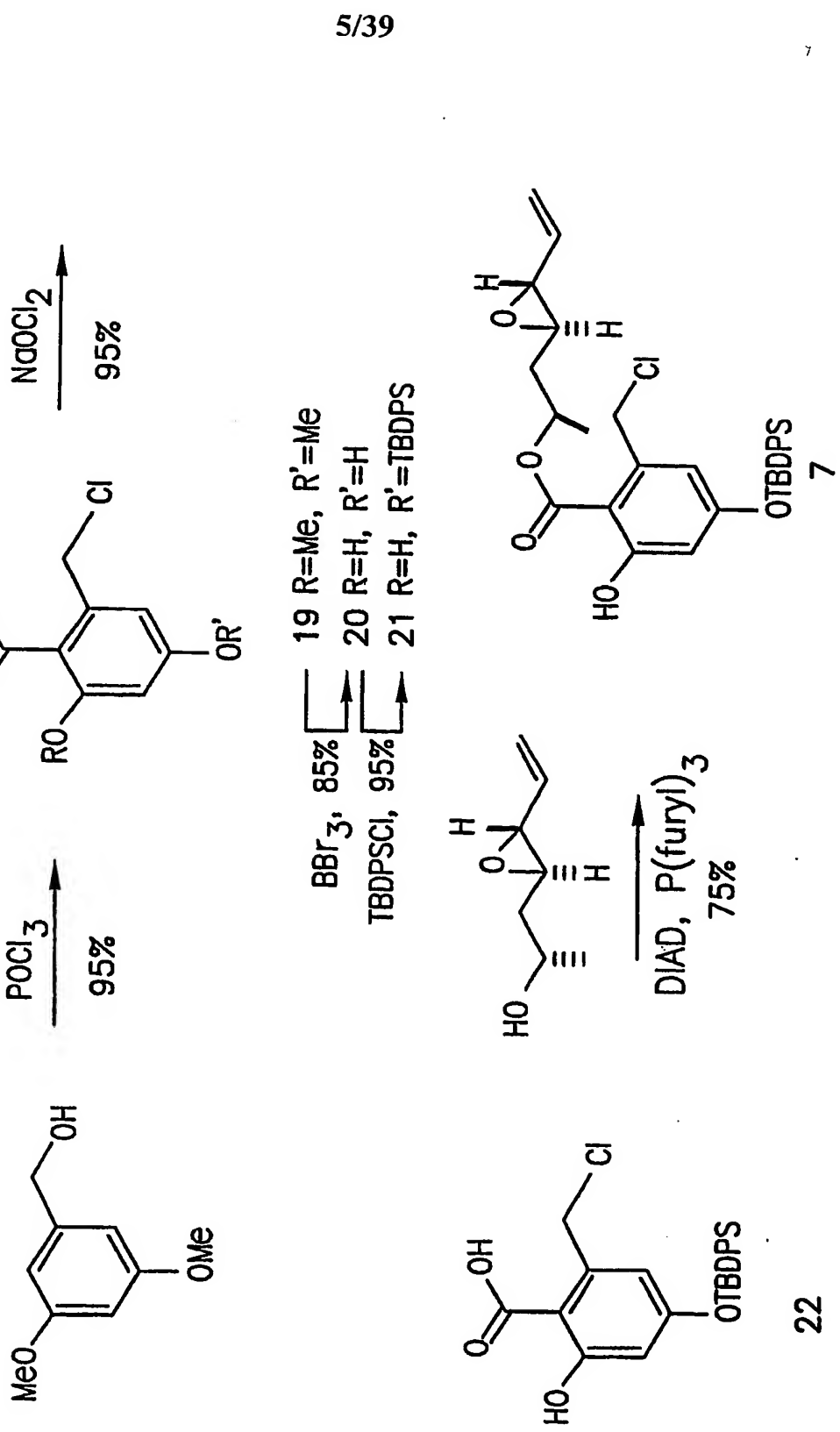
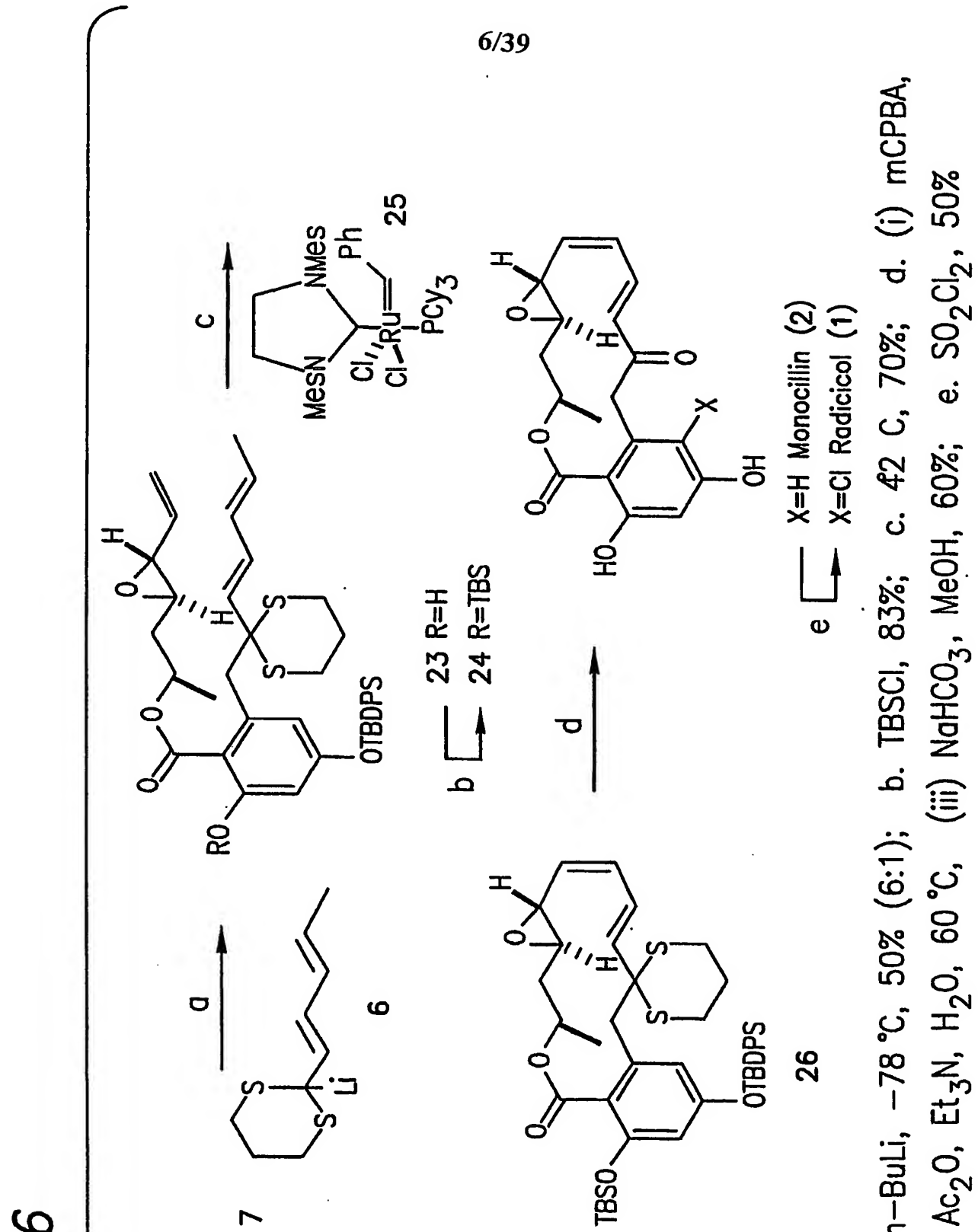

MeO
OMe
OH
POCl3
95%
RO
OR'
Cl
H
O
NaOCl2
95%
BBr3, 85%
TBDPSCl, 95%
19 R=Me, R'=Me
20 R=H, R'=H
21 R=H, R'=TBDPS
HO
OTBDPS
22
DIAD, P(furyl)3
75%
7

FIG. 6



a. n–BuLi, –78 °C, 50% (6:1); b. TBSCl, 83%; c. 42 C, 70%; d. (i) mCPBA, (ii) Ac_2O, Et_3N, H_2O, 60 °C, (iii) $NaHCO_3$, MeOH, 60%; e. SO_2Cl_2, 50%

FIG. 7

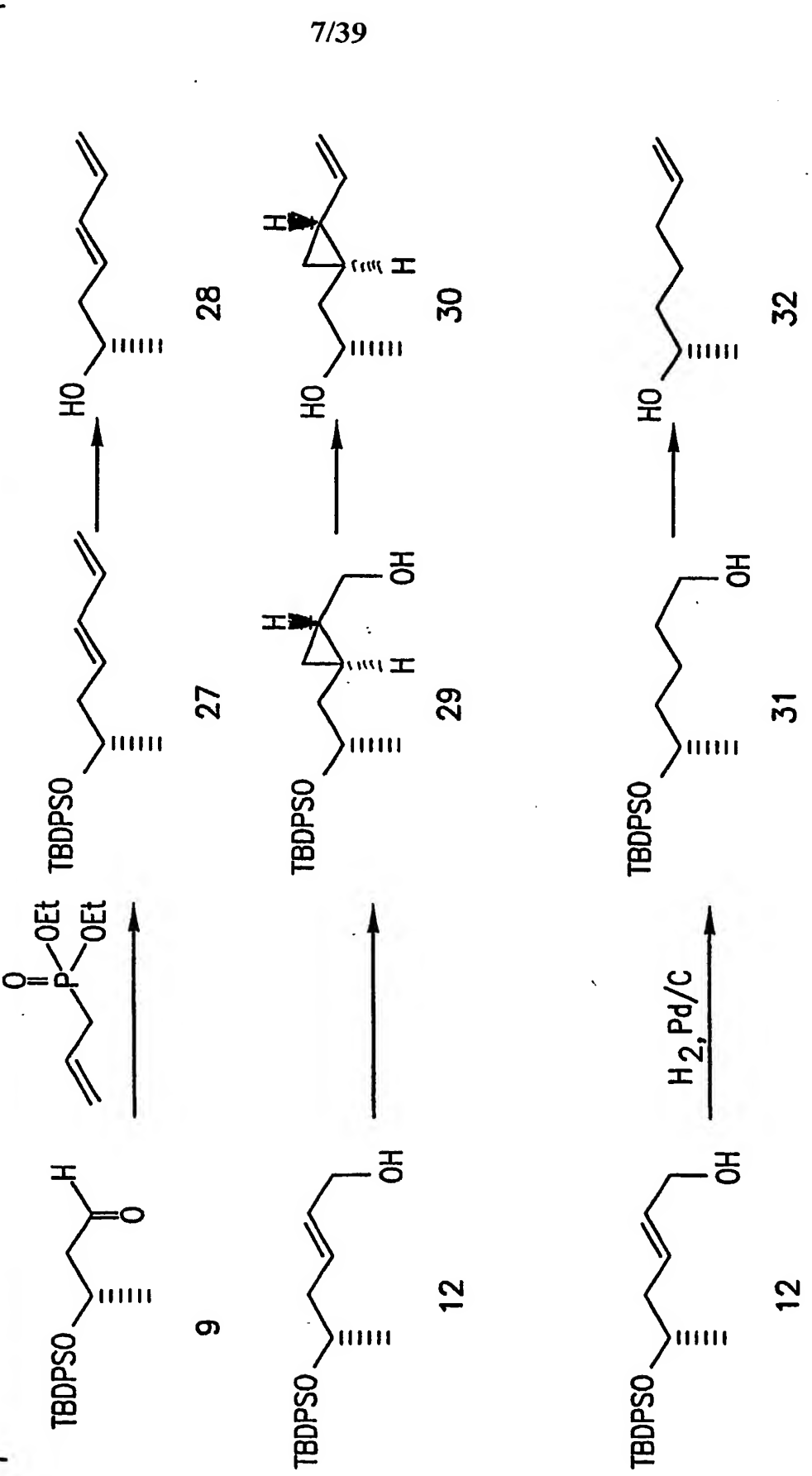

TBDPSO
H
O
9
O
P
OEt
OEt
TBDPSO
HO
27
28
TBDPSO
OH
12
H
OH
H
TBDPSO
HO
29
30
H2, Pd/C
TBDPSO
OH
12
TBDPSO
HO
31
32

FIG.8

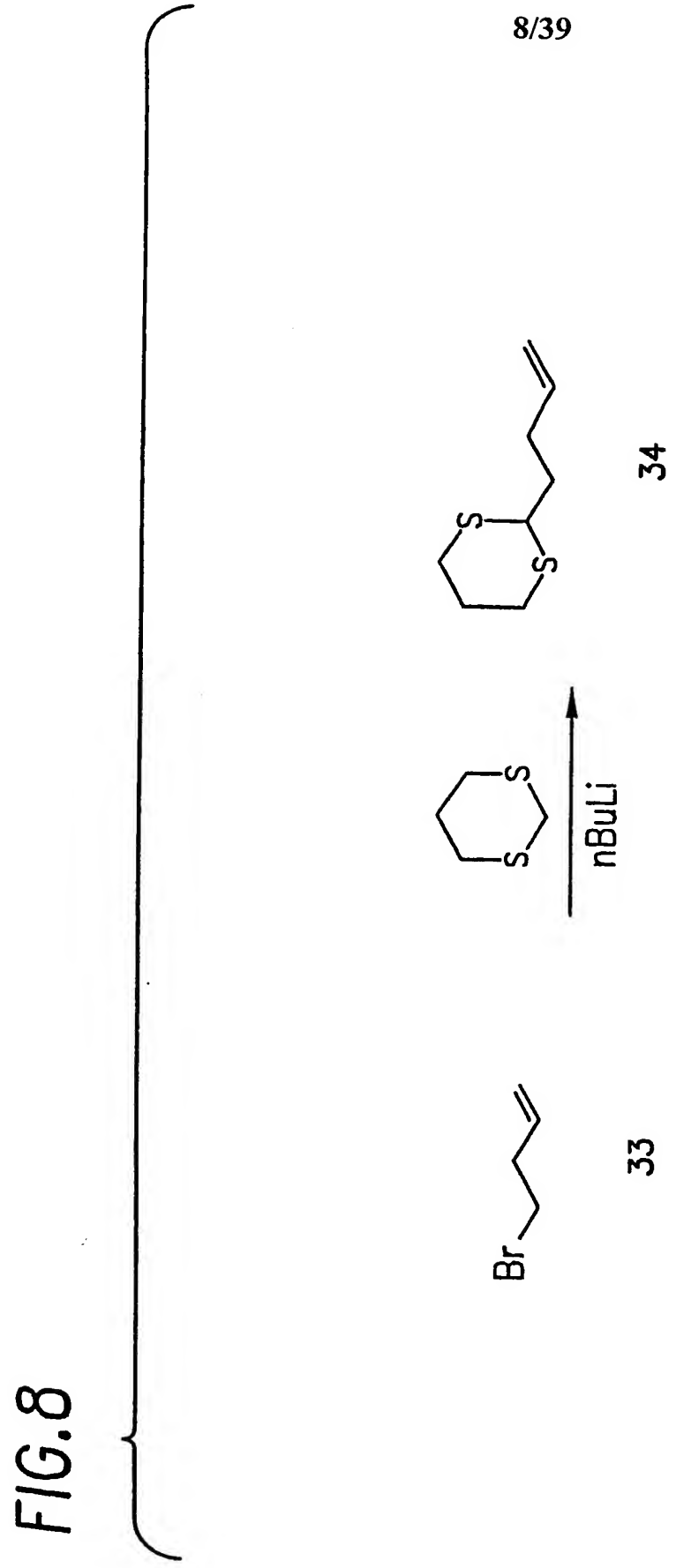


FIG.9

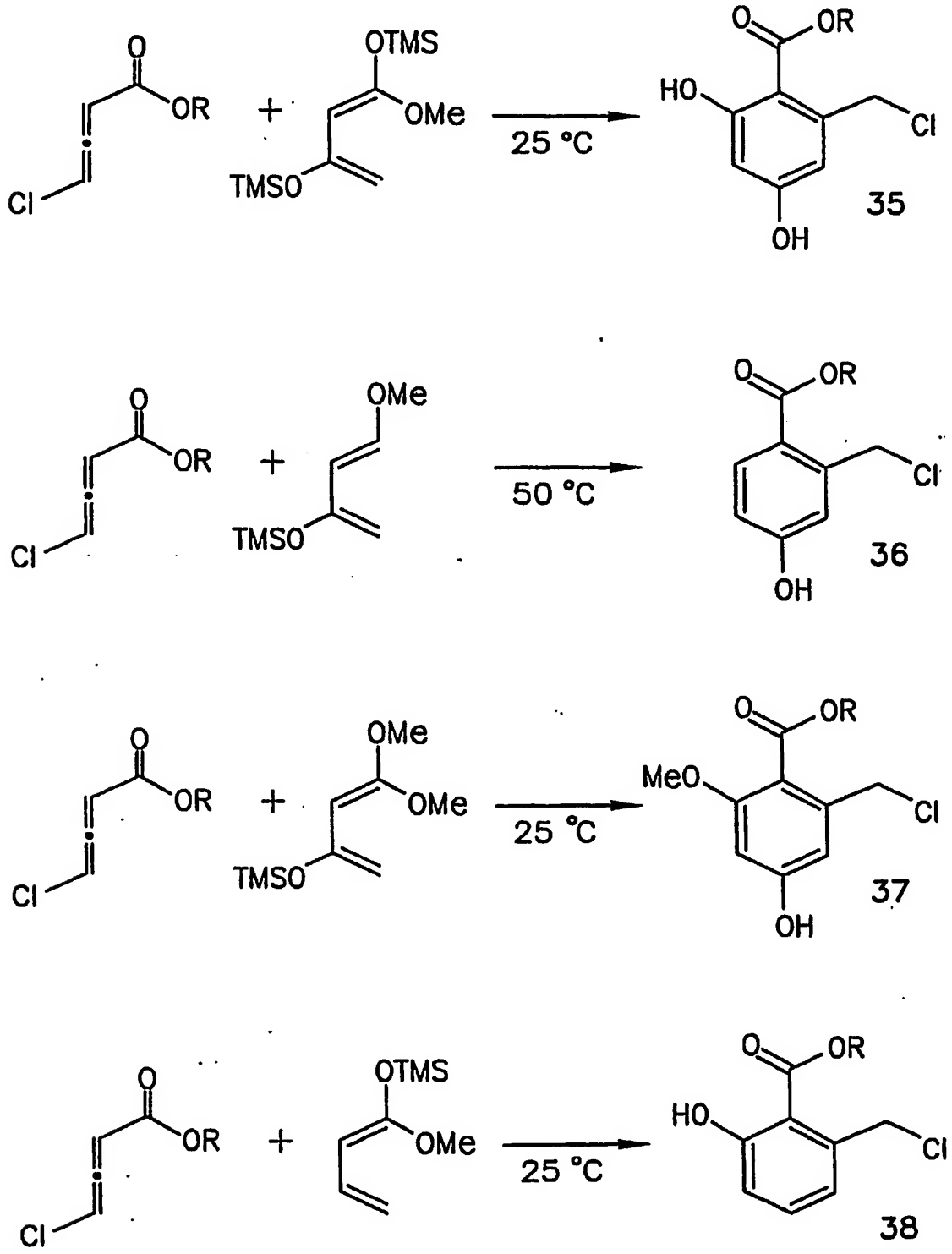
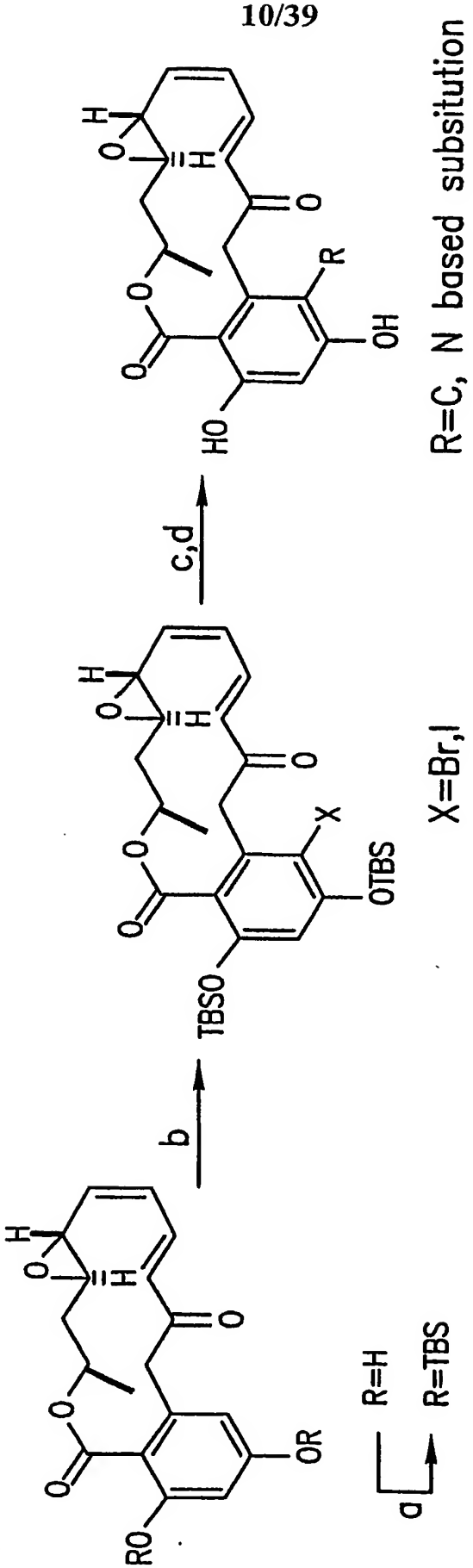
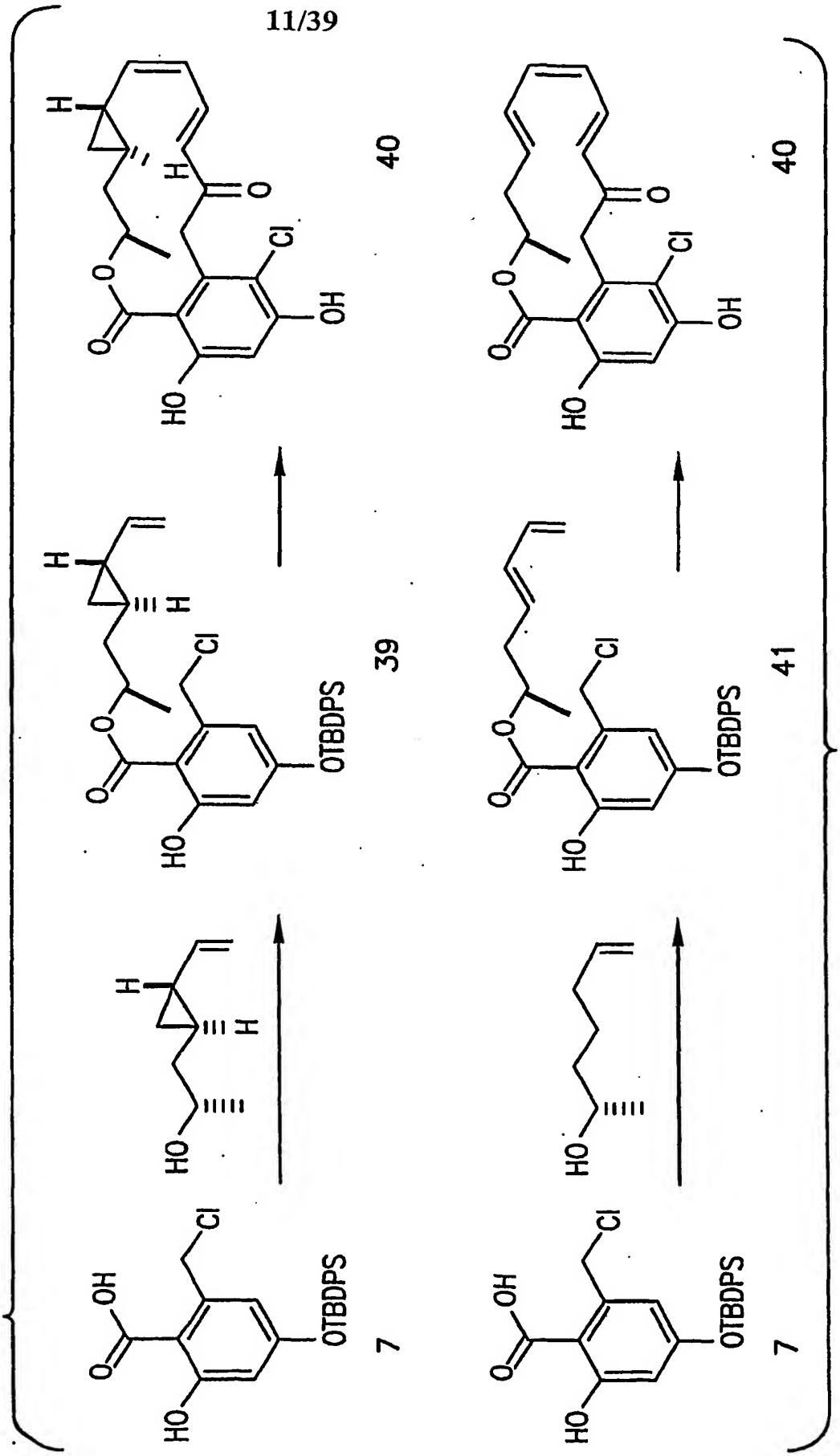


FIG. 10



a. TBSCl, pyridine; b. NIS or NBS, TsOH; c. $Pd(PPh)_3$, $RSnBu_3$, d. nBu_4NF

FIG. 11-1



TO FIG. 11-2

FIG. 11-2

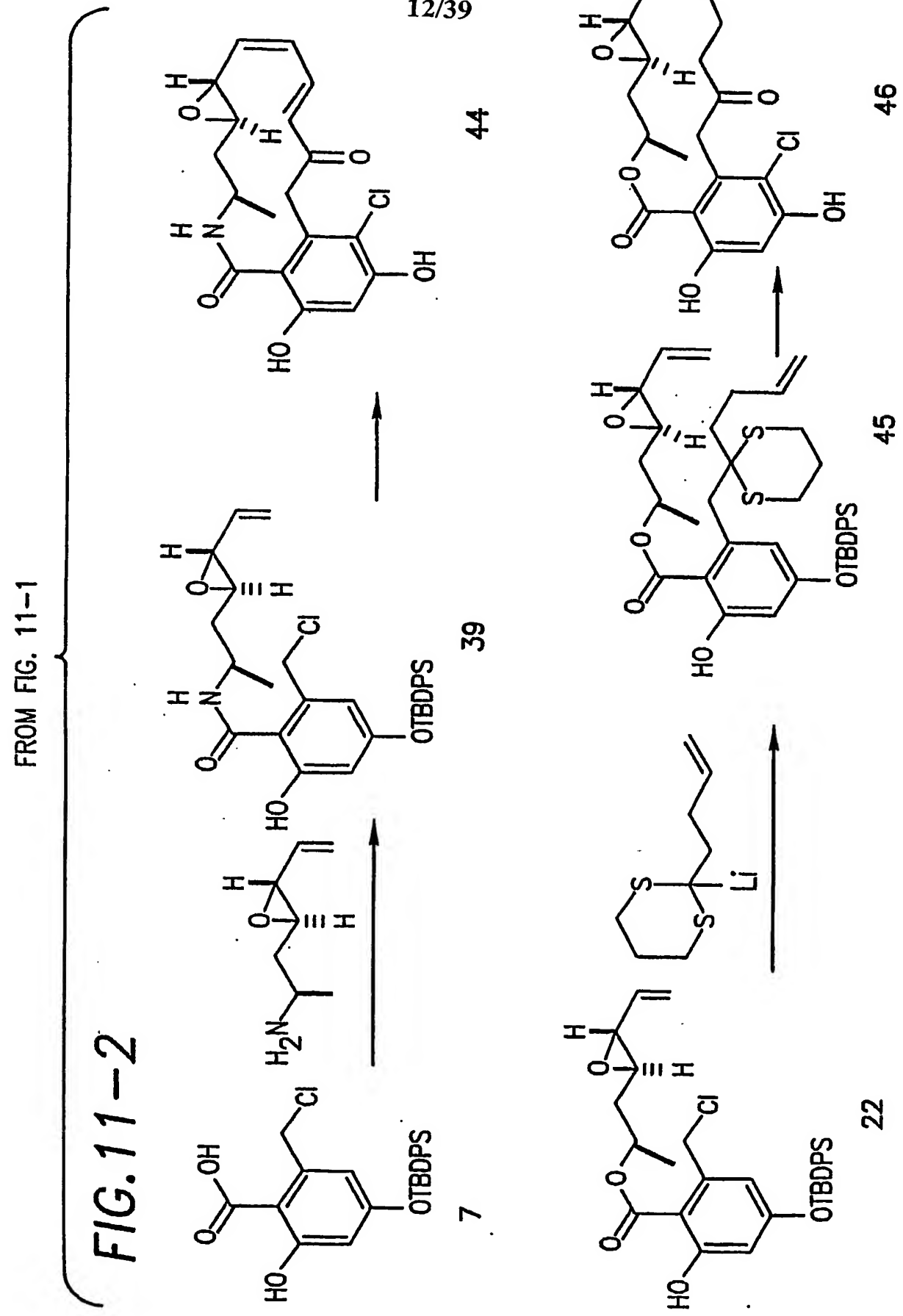
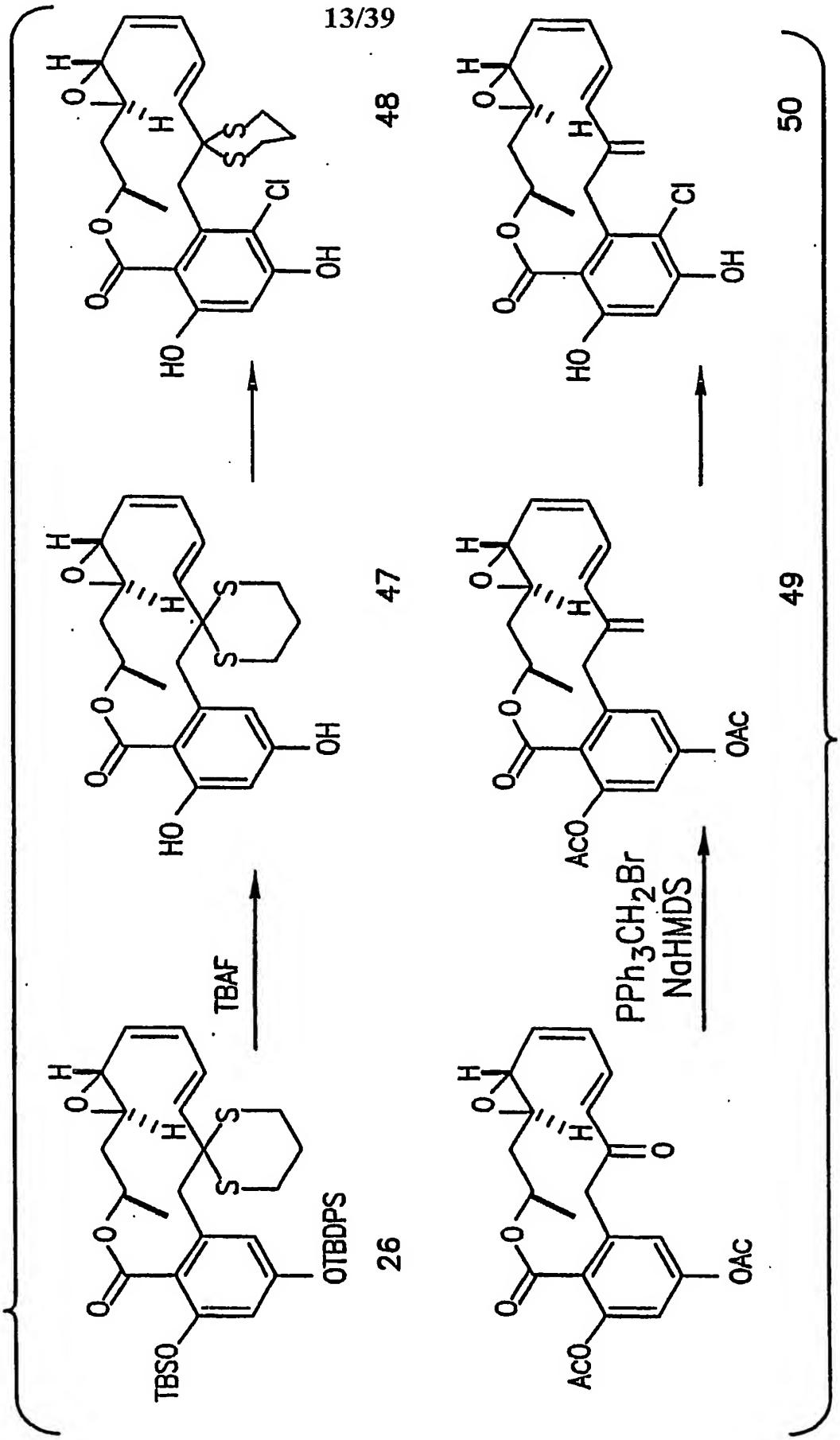


FIG. 12-1



TO FIG. 12-2

FIG. 12-2

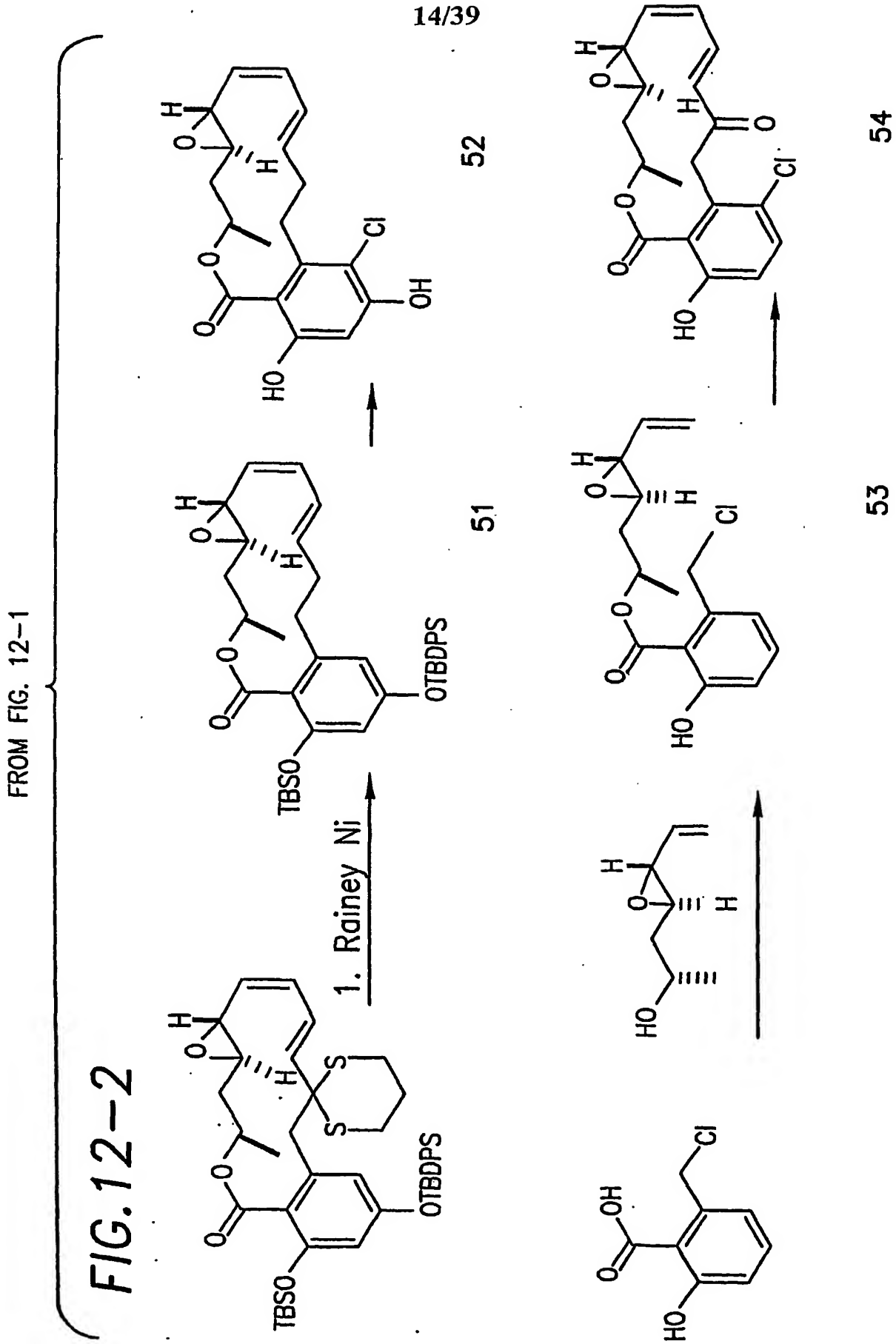
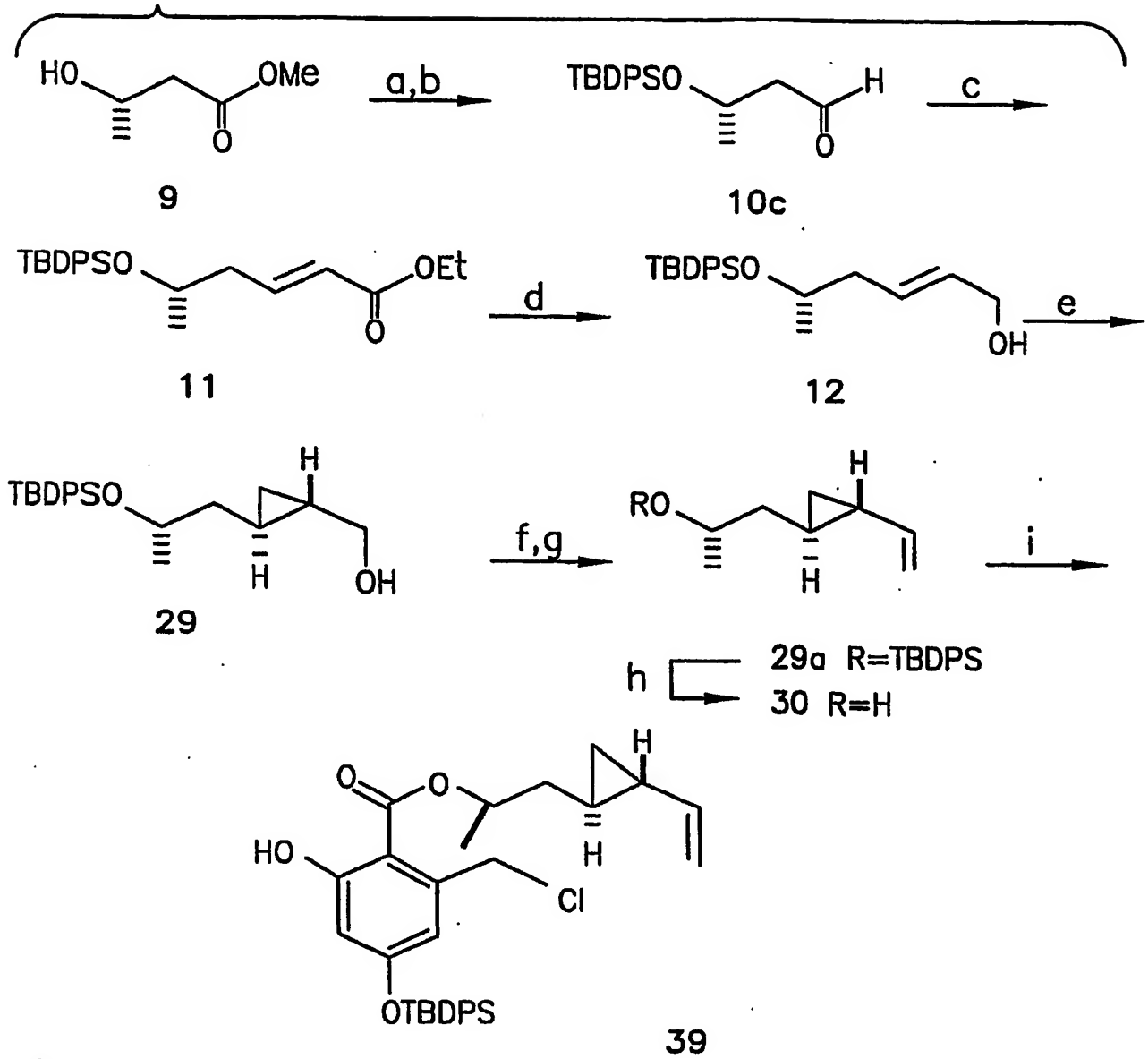
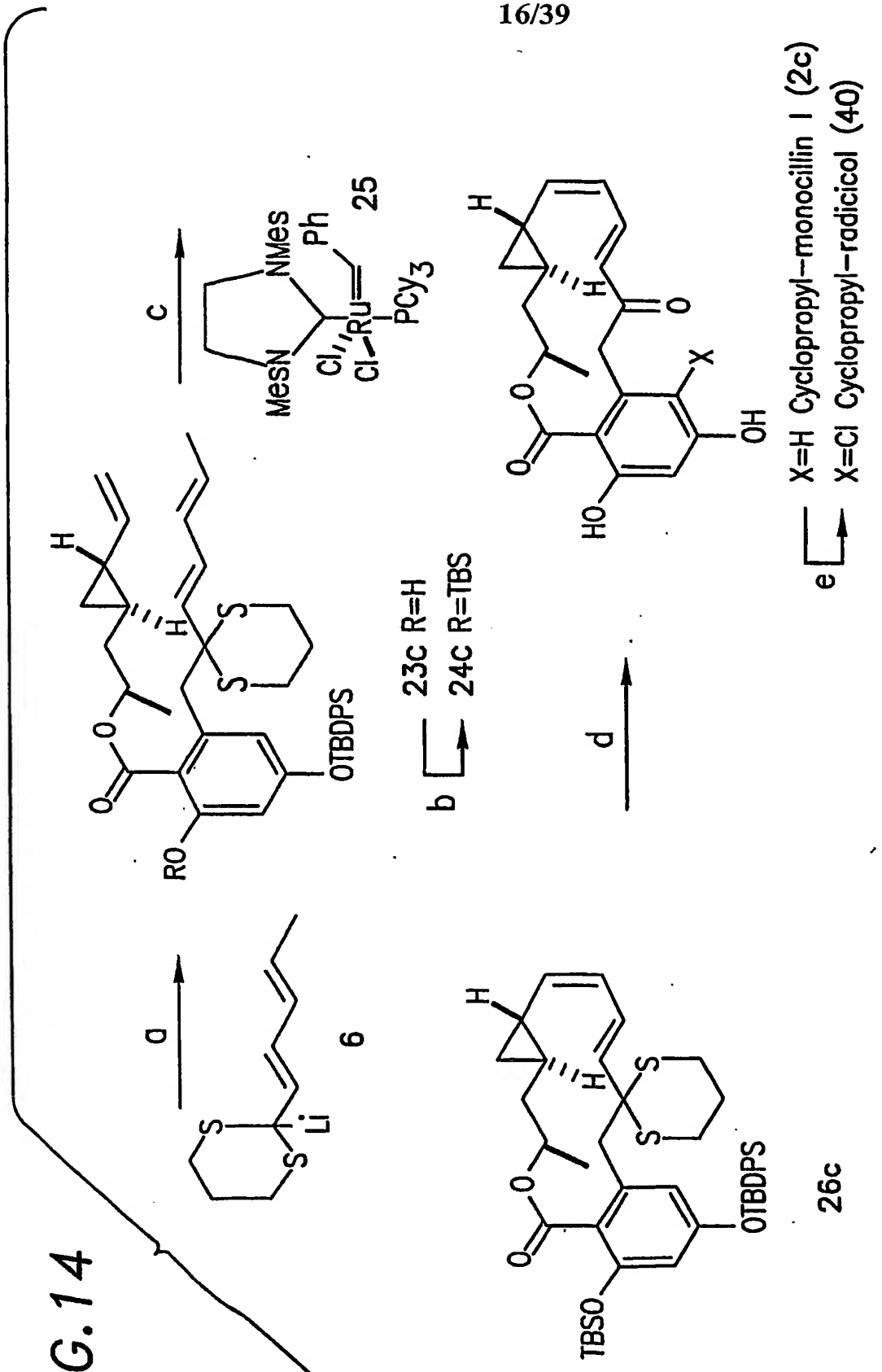


FIG. 13



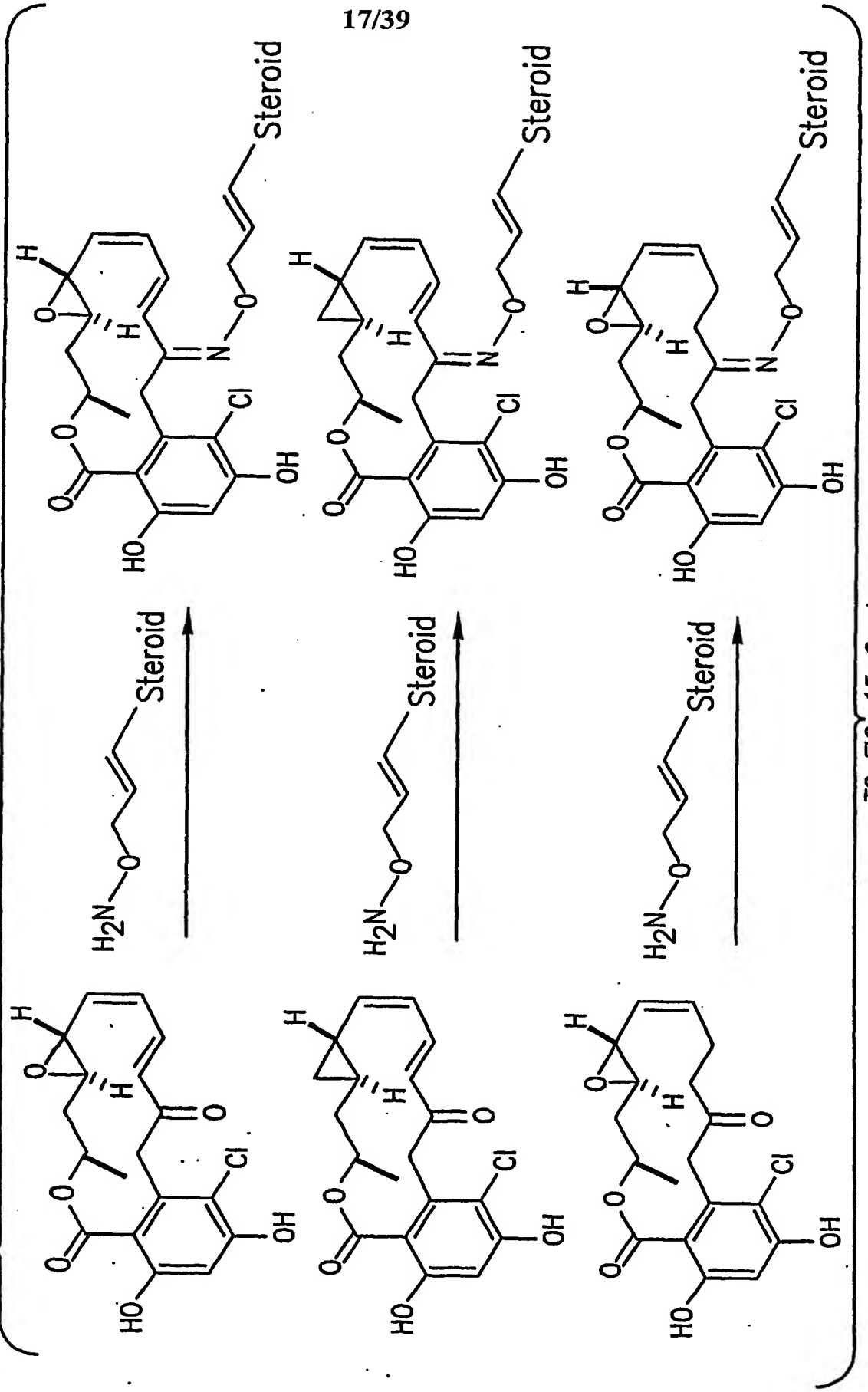
[a](a) TBDPSCl, imid.,>95%; (b) DIBAL–H,–78 °C, 92%; (c) LiCl, DIPEA $(EtO)_2P(O)CH_2CO_2Et$, 95%; (d) DIBAL–H –20 °C, 96%; (e) (+)–tetramethyltartaricacid diamide–BBu, Et_2Zn, CH_2I_2, 9 >95% ee; (f) SO_3*pyridine, Et_3N, DMSO, 90%; (g) Ph_3 PCH NaHMDS, 0 °C, 82%; (h) TBAF, 89%; (i) 7, $P(furyl)_3$, DIA benzene, 60%

FIG. 14



a. n-BuLi, −78 °C, 75% (3:1); b. TBSCl, 83%; c. 42 °C, 20%; d. (i) mCPBA, (ii) Ac_2O, Et_3N, H_2O, 60 °C, (iii) $NaHCO_3$, MeOH, 60%; e. SO_2Cl_2, 80%

FIG. 15-1



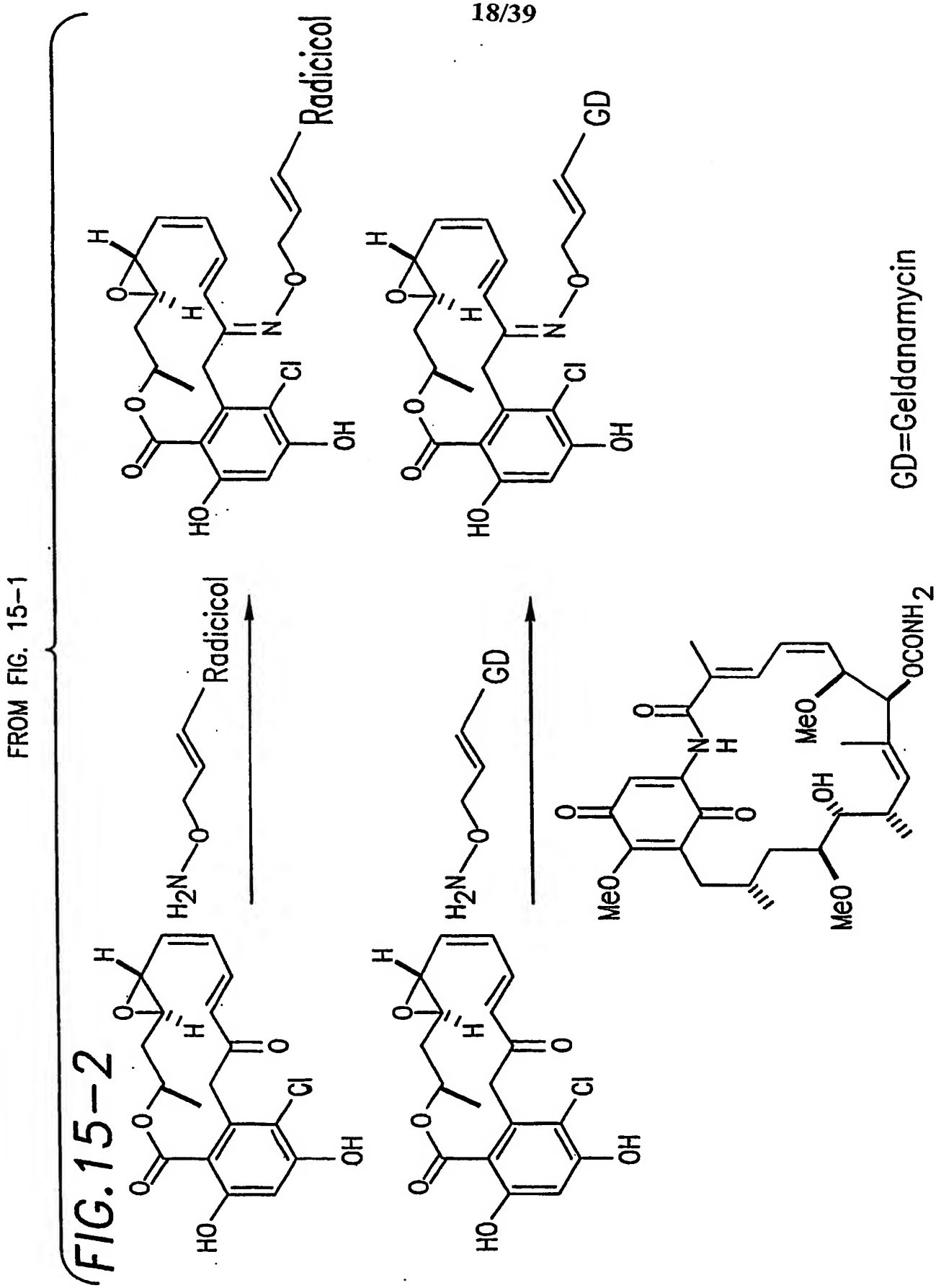

FROM FIG. 15-1
Radicicol
GD
H_2N
O
H
Cl
OH
HO
N
MeO
$OCONH_2$
GD=Geldanamycin

FIG. 15-2

FIG. 16-1

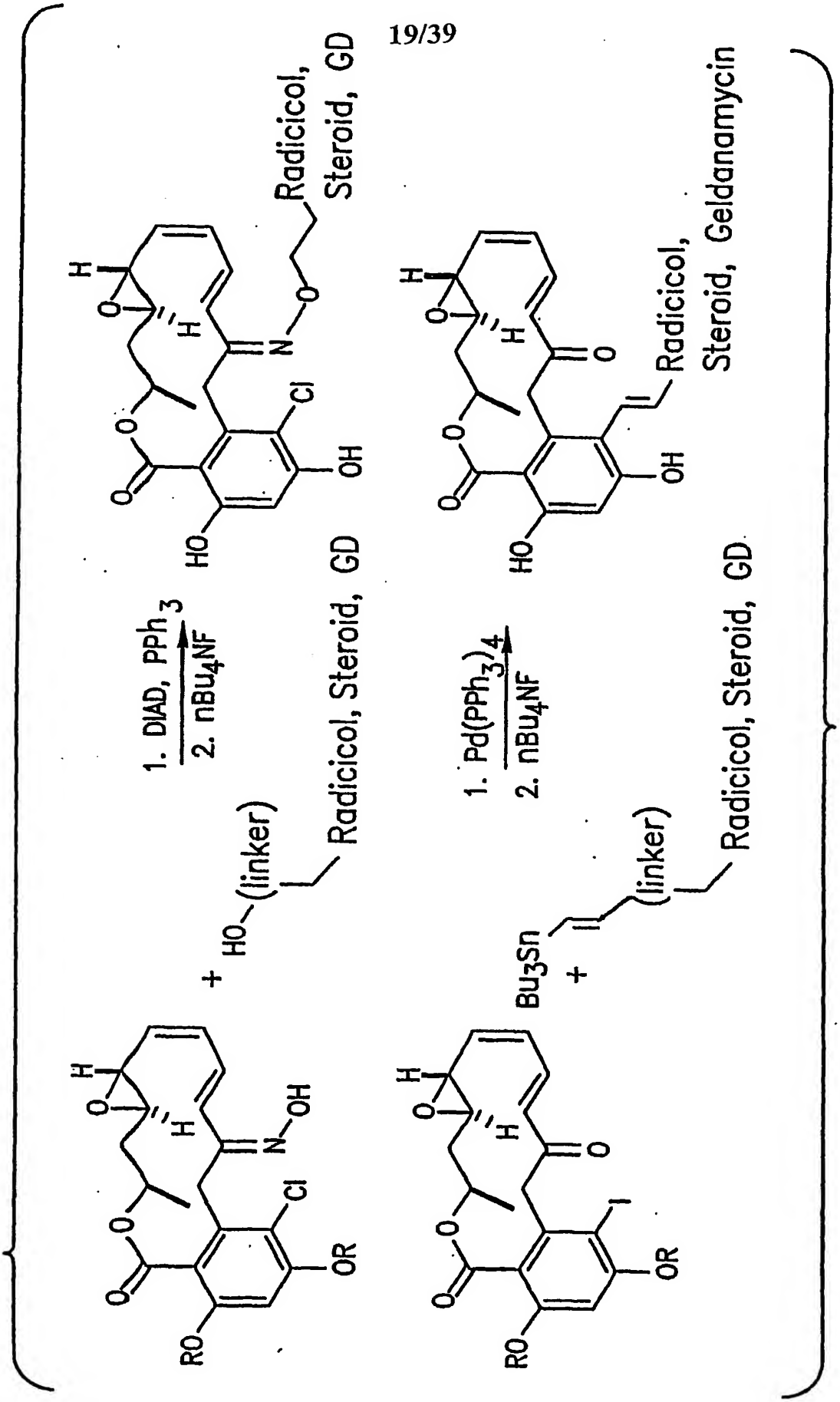

1. DIAD, PPh3
2. nBu4NF
HO—(linker)—Radicicol, Steroid, GD
Radicicol, Steroid, GD
1. Pd(PPh3)4
2. nBu4NF
Bu3Sn—(linker)—Radicicol, Steroid, GD
Radicicol, Steroid, Geldanamycin
TO FIG. 16-2

FIG. 16-2

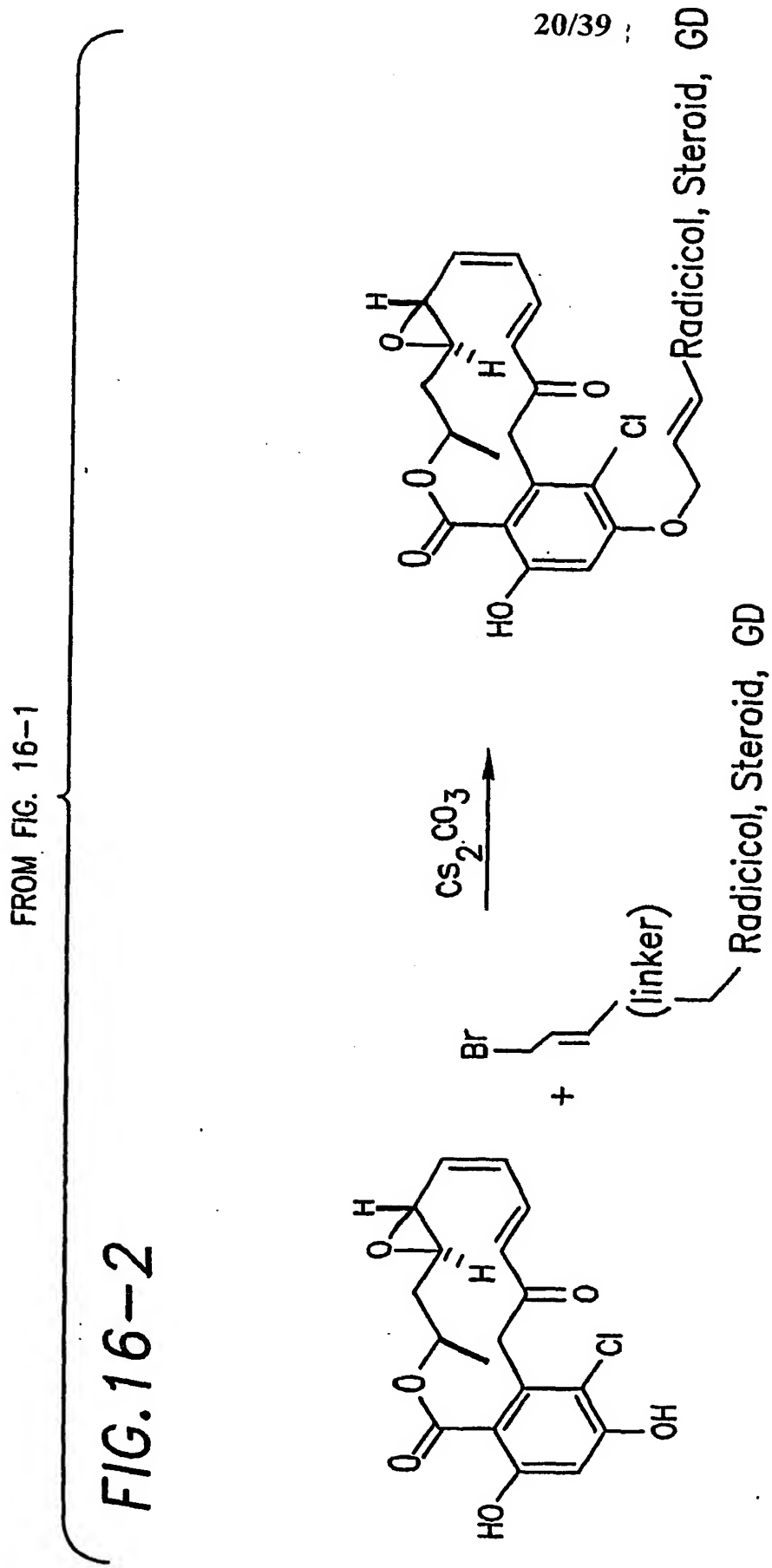

FROM FIG. 16-1
Cs2CO3
Br
(linker)
Radicicol, Steroid, GD
Radicicol, Steroid, GD
HO
OH
Cl
O
H

FIG. 17–1

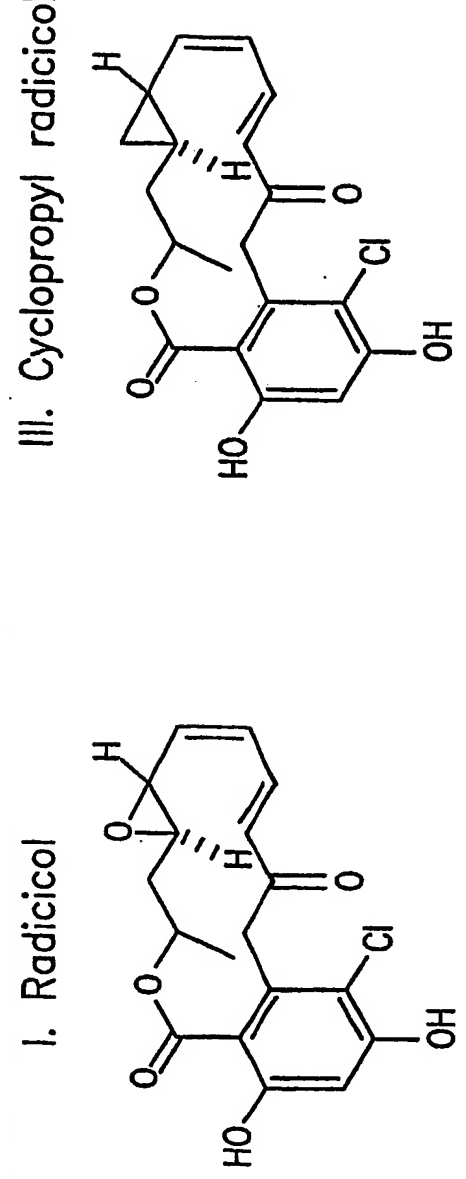

I. Radicicol
II. Monocillin I
III. Cyclopropyl radicicol
IV. Cyclopropyl monocillin
TO FIG. 17–2

FIG. 17-2

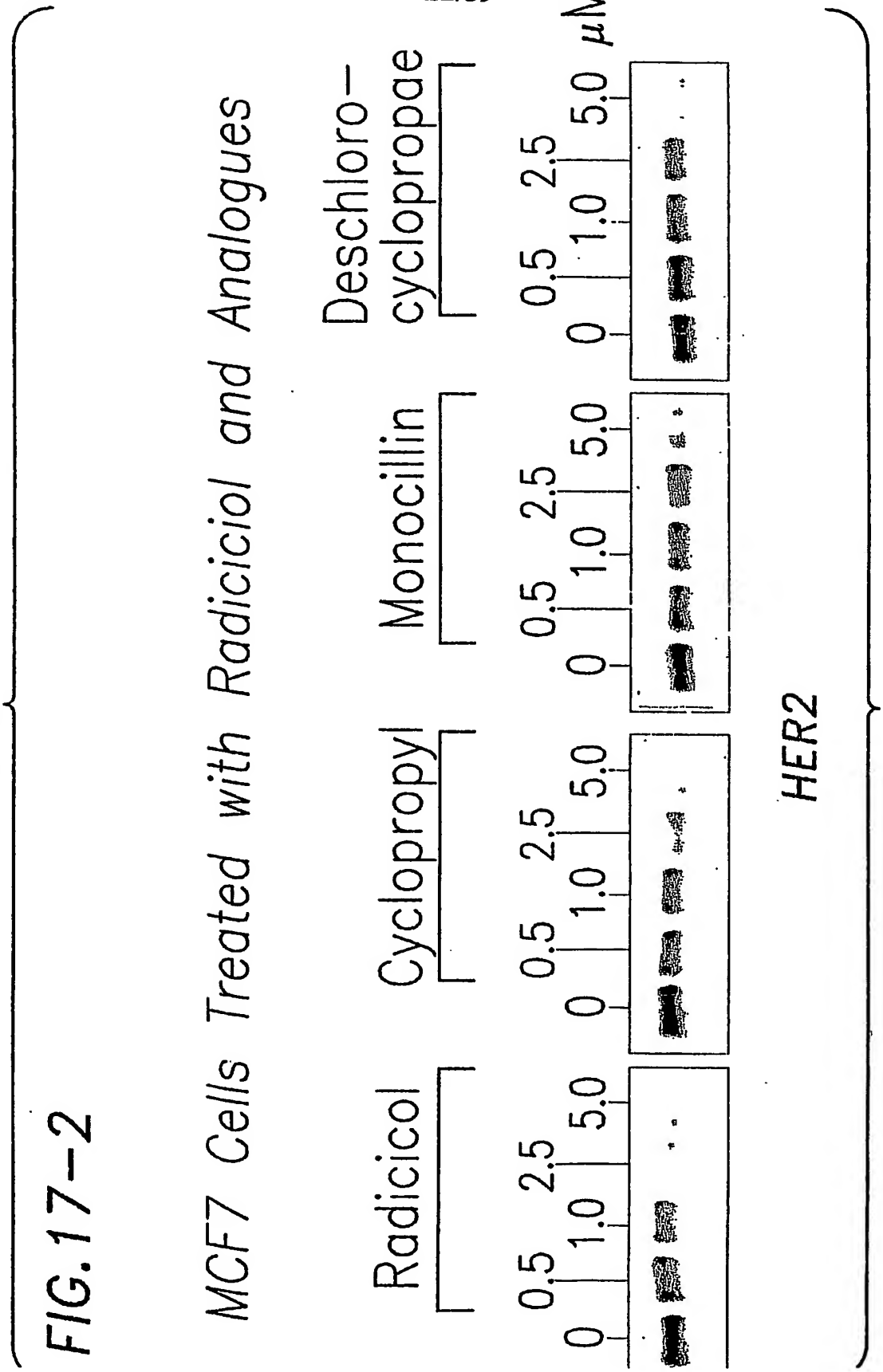
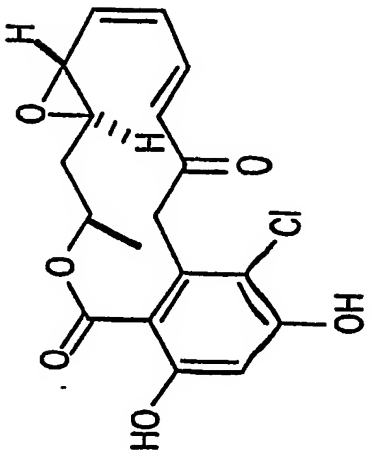

FROM FIG. 17-1
MCF7 Cells Treated with Radicicol and Analogues
Radicicol
Cyclopropyl
Monocillin
Deschloro-cyclopropae
0 0.5 1.0 2.5 5.0 μM
HER2
TO FIG. 17-3

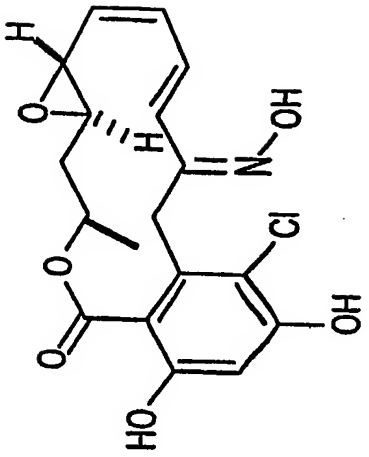
FIG. 17–3

FROM FIG. 17–2

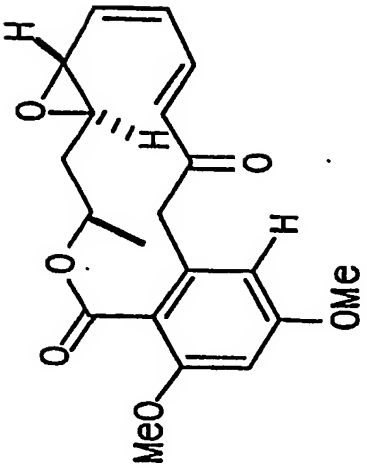
I. Radicicol



VII. Radicicol Oxime



V. Dimethyl Monocillin I



VI. Dimethyl Radicicol

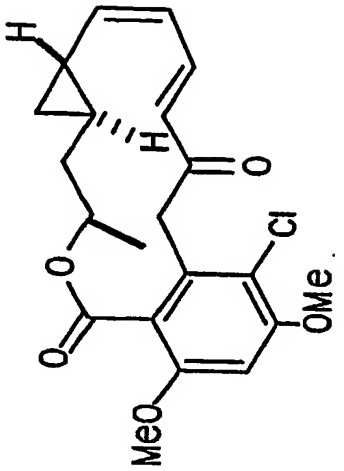
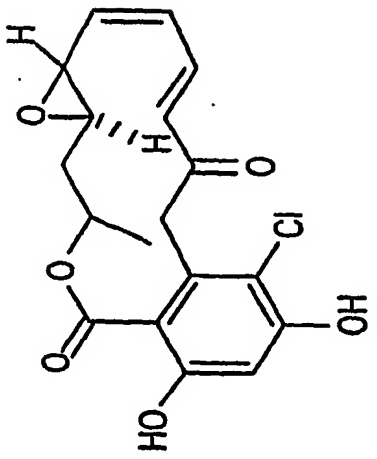
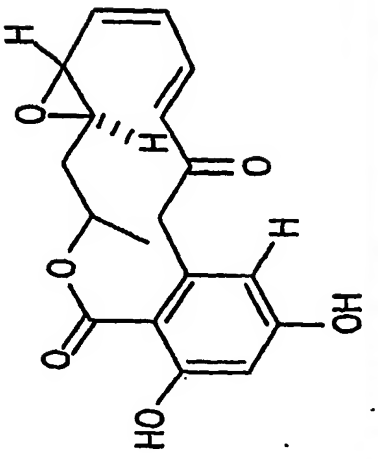


FIG. 18–1

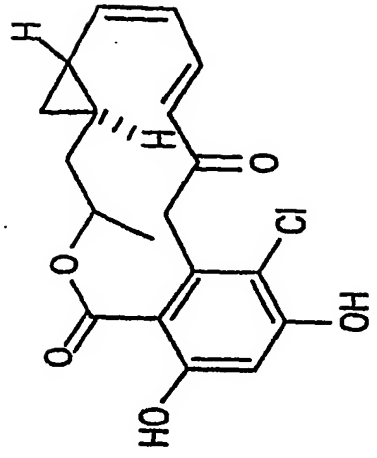
I. Radicicol



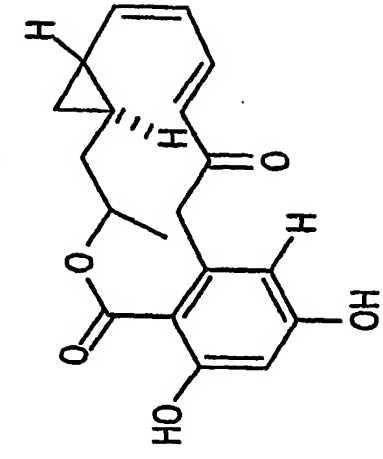
II. Monocillin I



III. Cyclopropyl radicicol



IV. Cyclopropyl monocillin

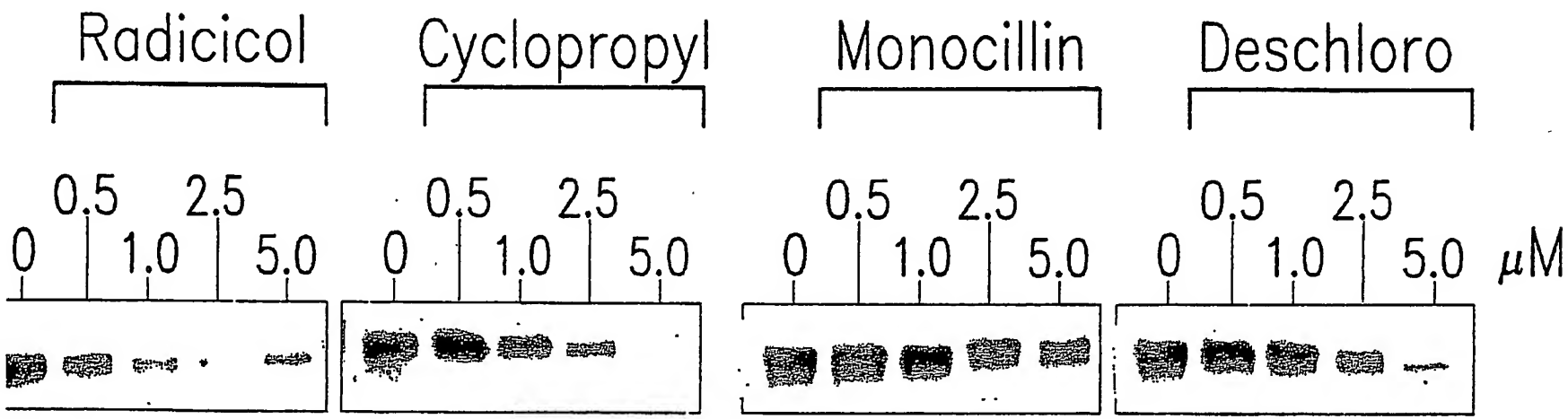


TO FIG. 18–2

FROM FIG. 18-1

FIG. 18-2

BT474 Cells Treated with Novel Radiciciols (24hrs.)



HER2

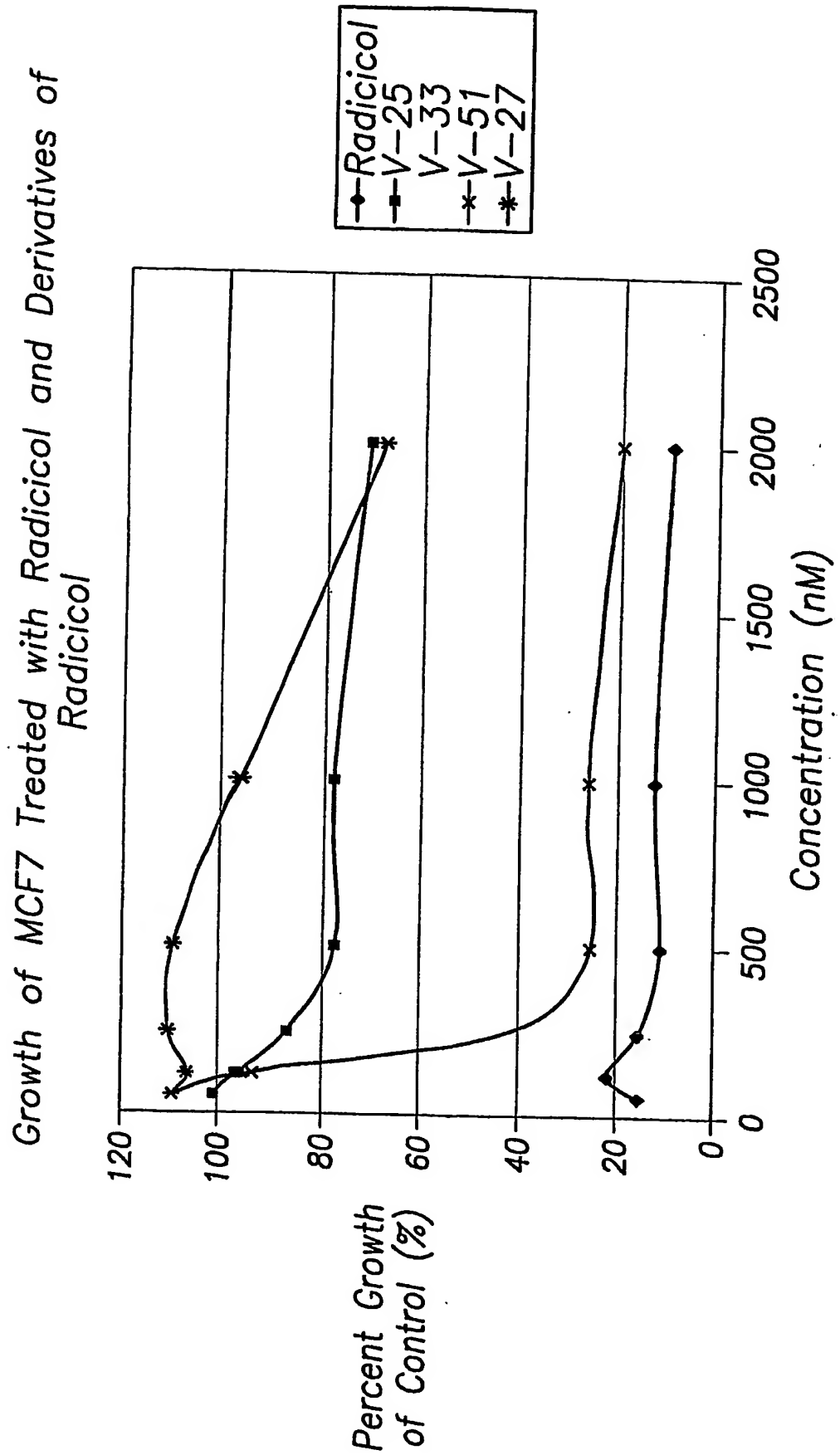

Growth of MCF7 Treated with Radicicol and Derivatives of Radicicol
Radicicol
V-25
V-33
V-51
V-27
Percent Growth of Control (%)
120
100
80
60
40
20
0
0
500
1000
1500
2000
2500
Concentration (nM)

FIG. 19

FIG.20

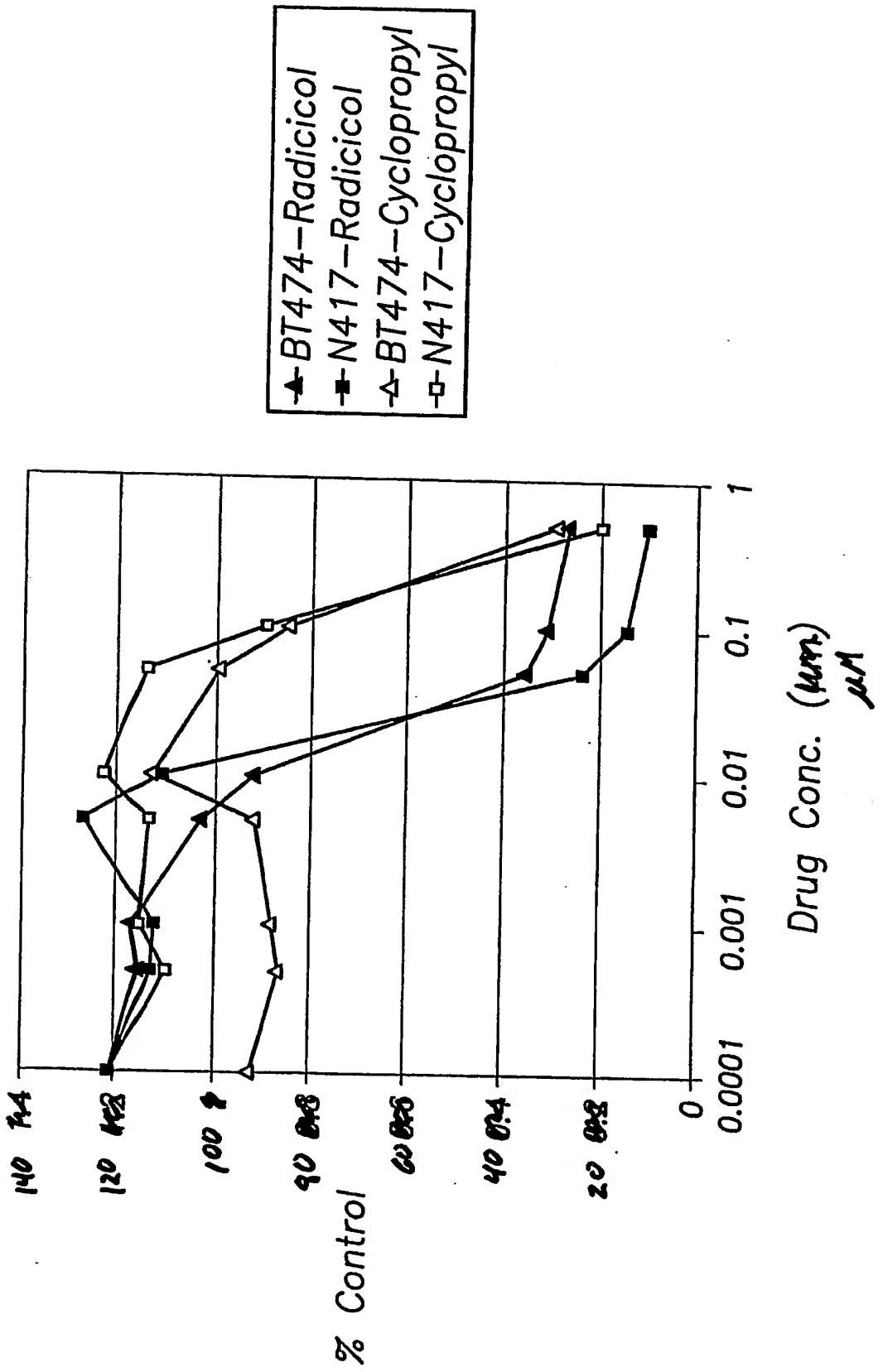
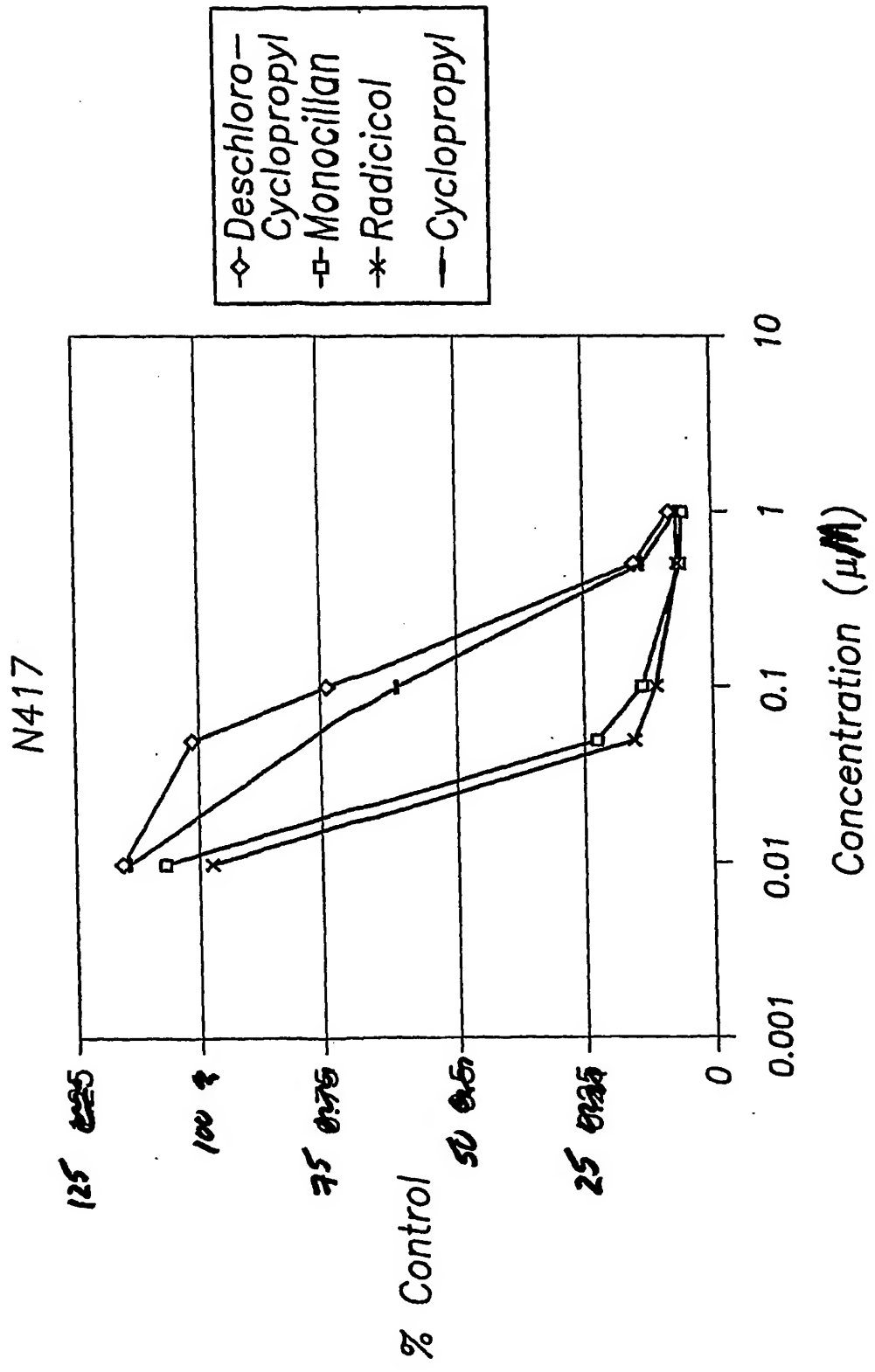

BT474–Radicicol
N417–Radicicol
BT474–Cyclopropyl
N417–Cyclopropyl
% Control
140
120
100
90
80
60
40
20
0
0.0001
0.001
0.01
0.1
1
Drug Conc. (μM)

FIG.21


Deschloro-Cyclopropyl
Monocillan
Radicicol
Cyclopropyl
N417
% Control
125
100
75
50
25
0
0.001
0.01
0.1
1
10
Concentration (μM)

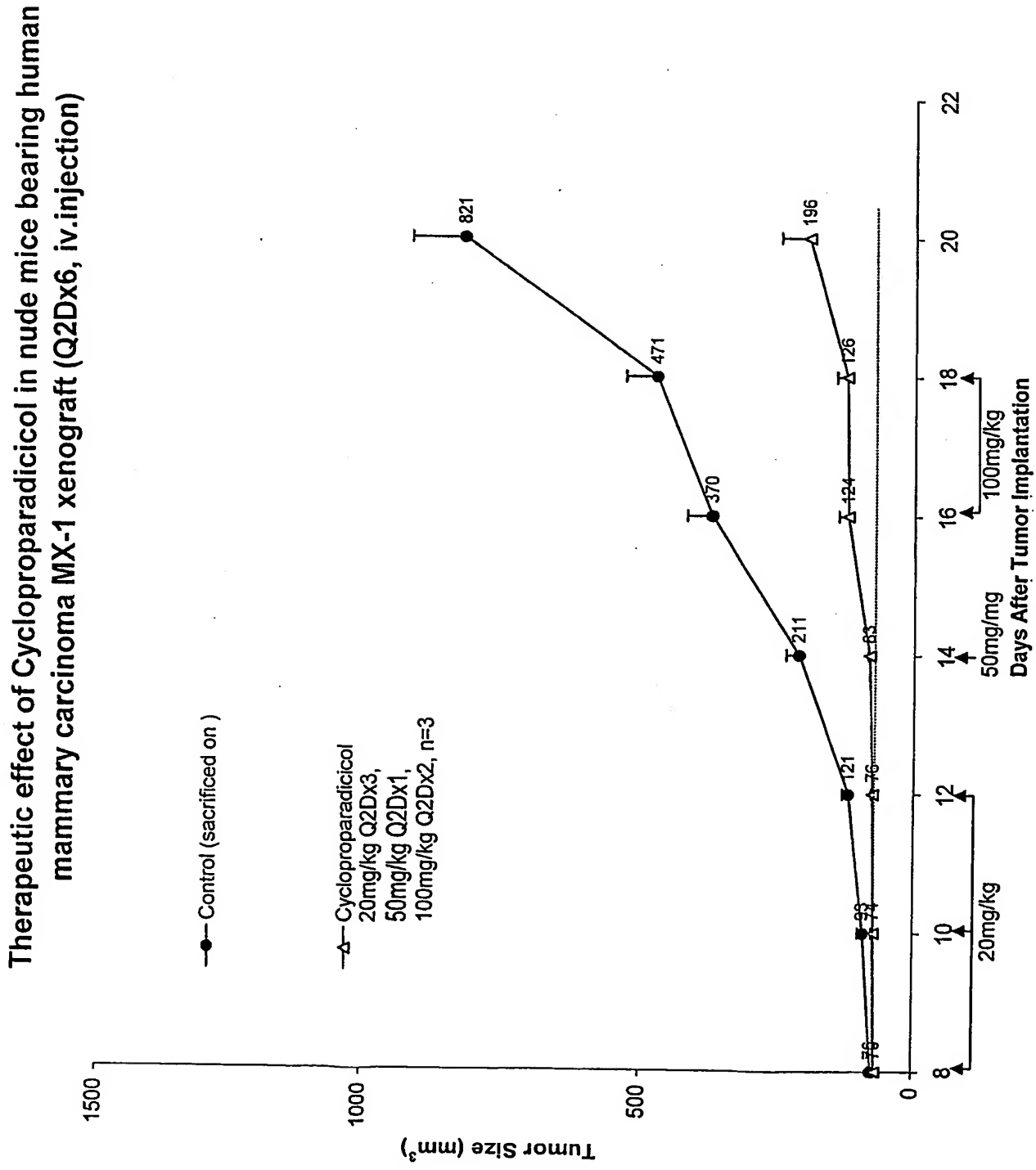

Therapeutic effect of Cyclopropradicicol in nude mice bearing human mammary carcinoma MX-1 xenograft (Q2Dx6, iv.injection)
Control (sacrificed on)
Cyclopropradicicol 20mg/kg Q2Dx3, 50mg/kg Q2Dx1, 100mg/kg Q2Dx2, n=3
Tumor Size (mm³)
1500
1000
500
0
821
471
370
211
121
76
196
126
124
83
76
93
74
8
10
12
14
16
18
20
22
20mg/kg
50mg/mg
100mg/kg
Days After Tumor Implantation

Fig. 22

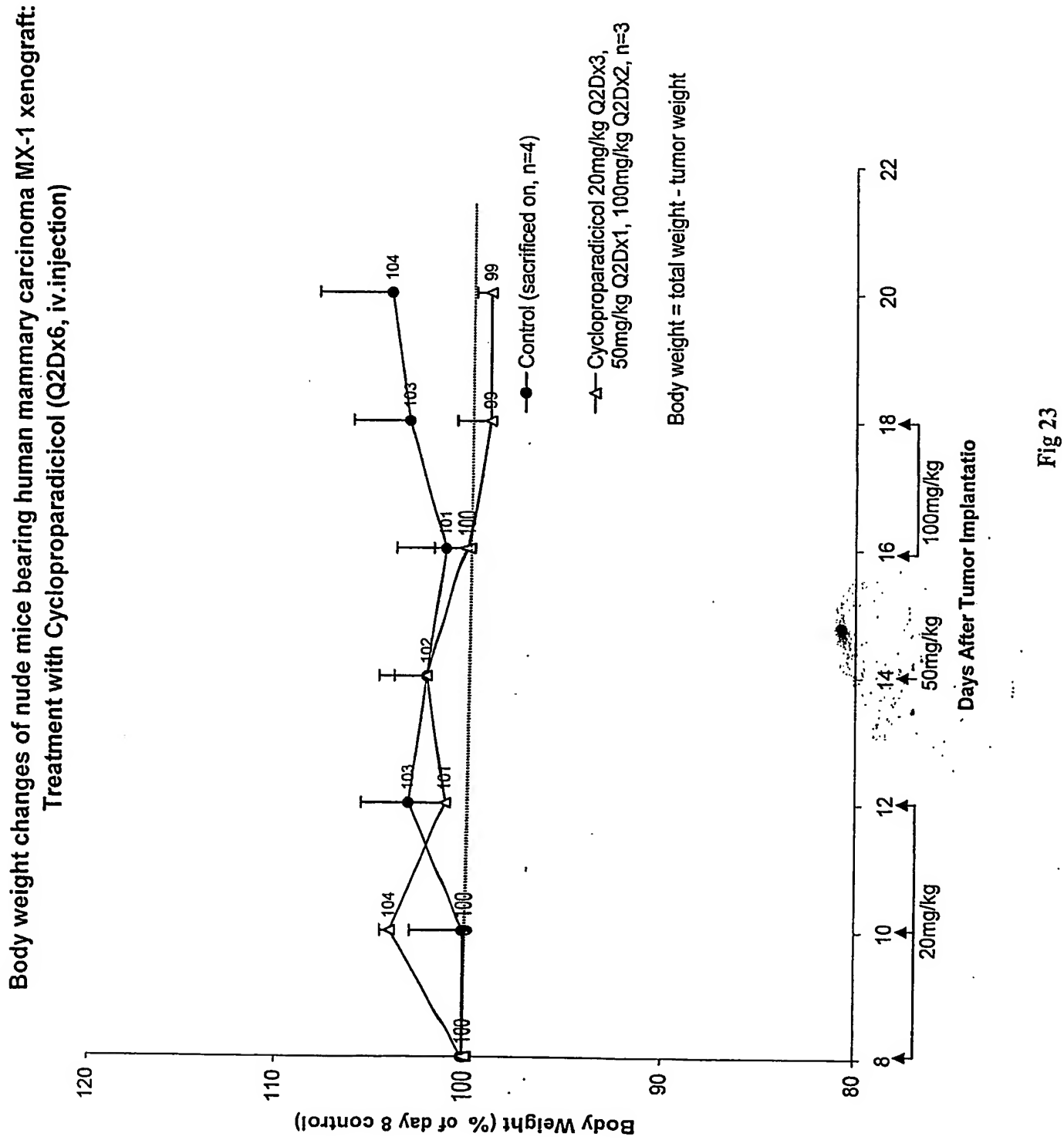

Body weight changes of nude mice bearing human mammary carcinoma MX-1 xenograft:
Treatment with Cycloproparadicicol (Q2Dx6, iv.injection)
Body Weight (% of day 8 control)
120
110
100
90
80
8
10
12
14
16
18
20
22
Days After Tumor Implantatio
20mg/kg
50mg/kg
100mg/kg
Control (sacrificed on, n=4)
Cycloproparadicicol 20mg/kg Q2Dx3,
50mg/kg Q2Dx1, 100mg/kg Q2Dx2, n=3
Body weight = total weight - tumor weight
104
103
101
100
99
102

Fig 23

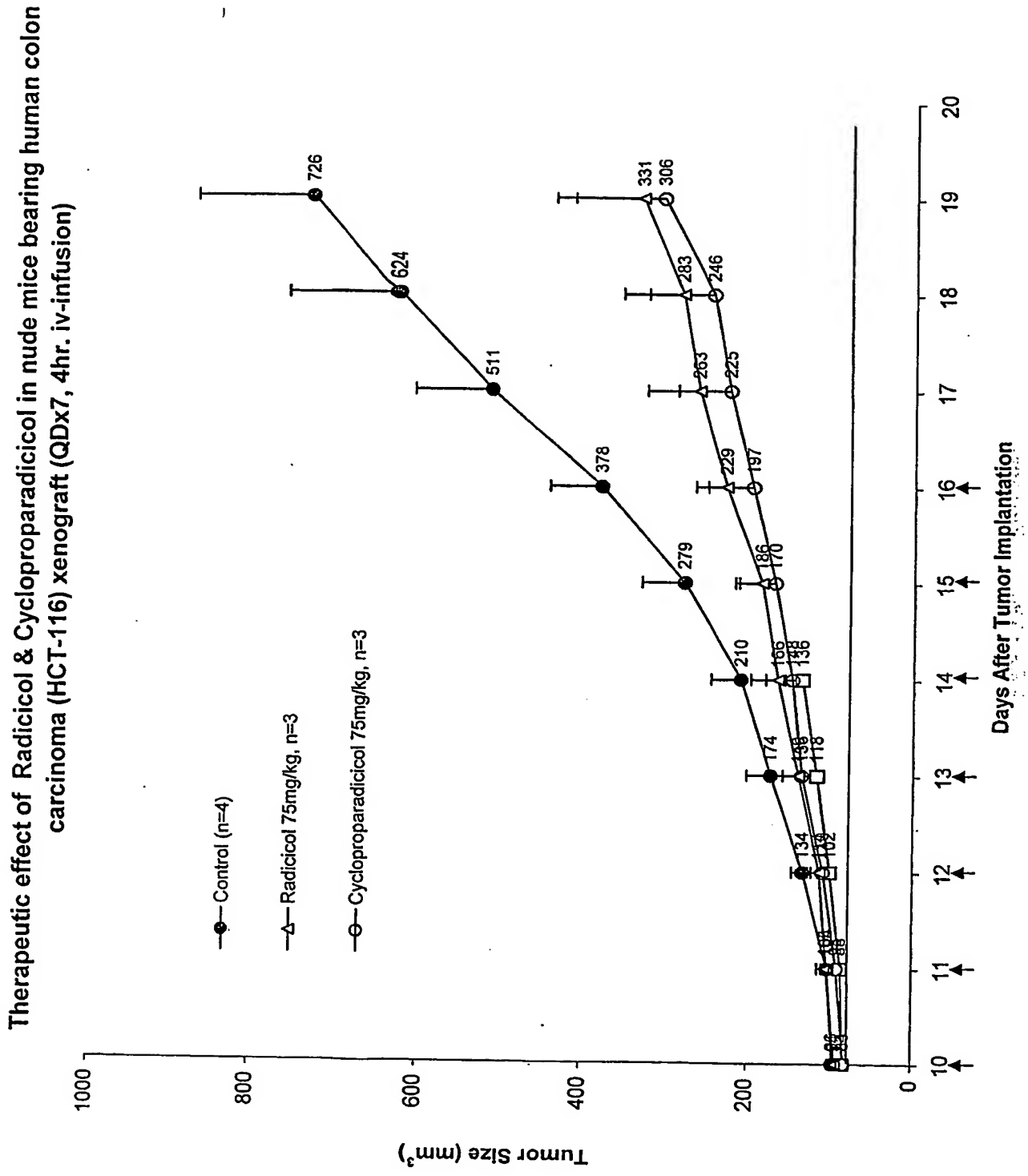

Therapeutic effect of Radicicol & Cyclopropparadicicol in nude mice bearing human colon carcinoma (HCT-116) xenograft (QDx7, 4hr. iv-infusion)
Control (n=4)
Radicicol 75mg/kg, n=3
Cyclopropparadicicol 75mg/kg, n=3
Tumor Size (mm³)
Days After Tumor Implantation
1000
800
600
400
200
0
10
11
12
13
14
15
16
17
18
19
20
726
624
511
378
279
210
174
134
331
306
283
246
263
225
229
197
186
170
166
138
136
118

Fig. 24

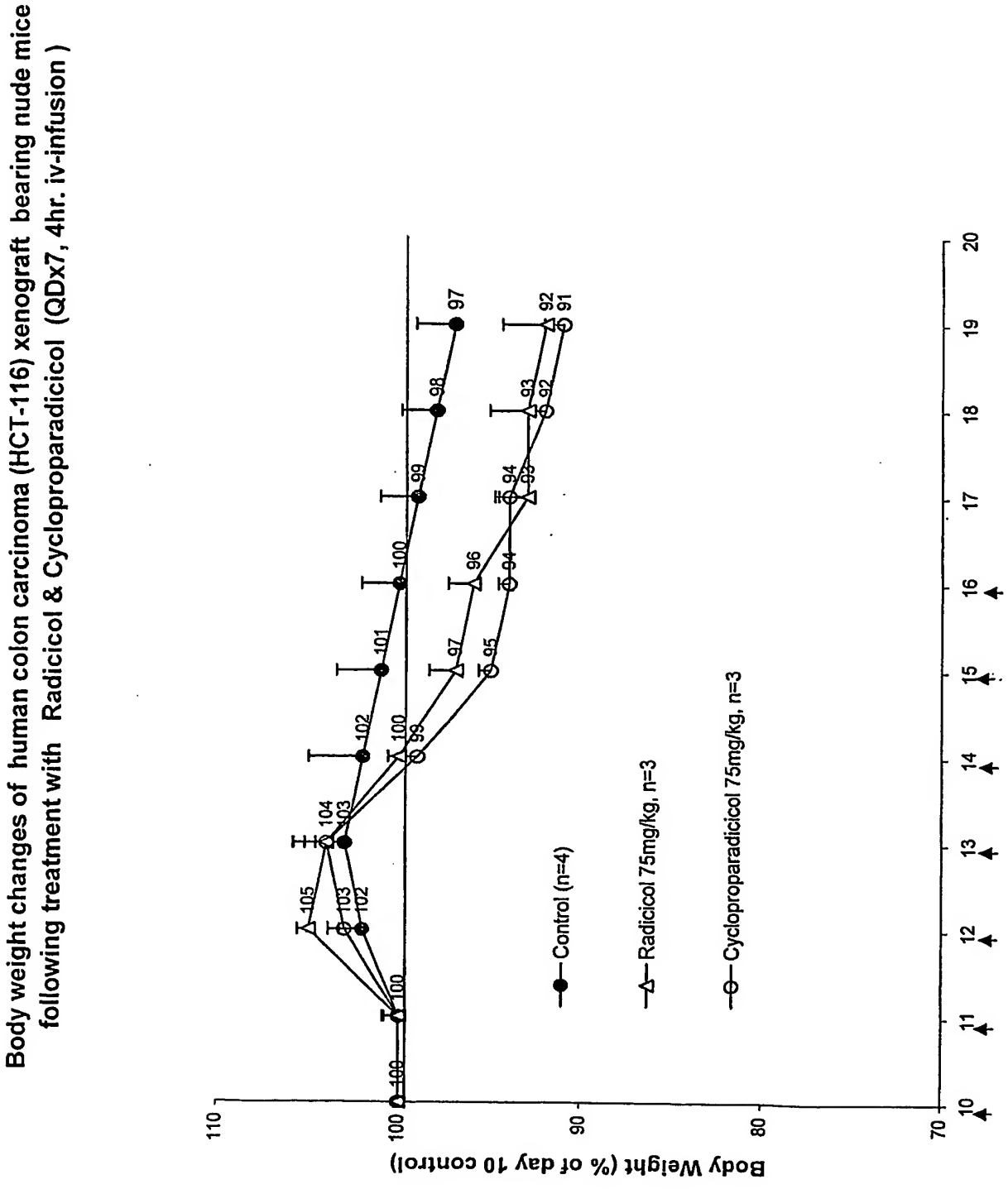

Body weight changes of human colon carcinoma (HCT-116) xenograft bearing nude mice following treatment with Radicicol & Cyclopropparadicicol (QDx7, 4hr. iv-infusion)
Body Weight (% of day 10 control)
Days After Tumor Implantation
Control (n=4)
Radicicol 75mg/kg, n=3
Cyclopropparadicicol 75mg/kg, n=3

Fig. 25

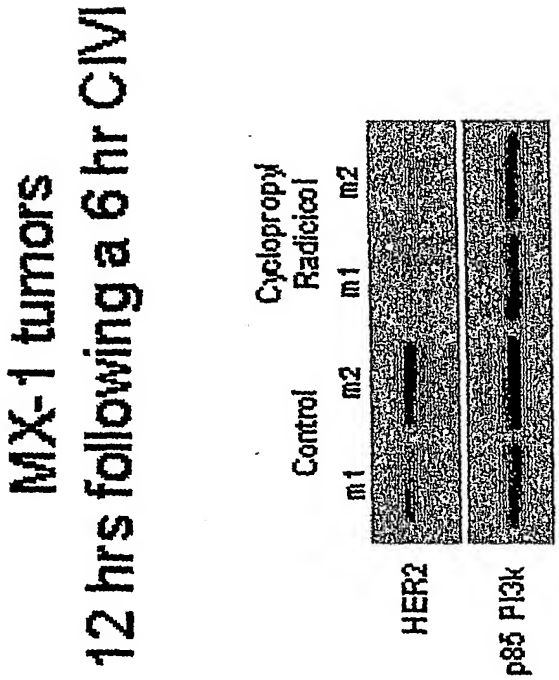

MX-1 tumors
12 hrs following a 6 hr CIVI
Control
Cyclopropyl Radicicol
m1
m2
m1
m2
HER2
p85 PI3k

Fig. 26

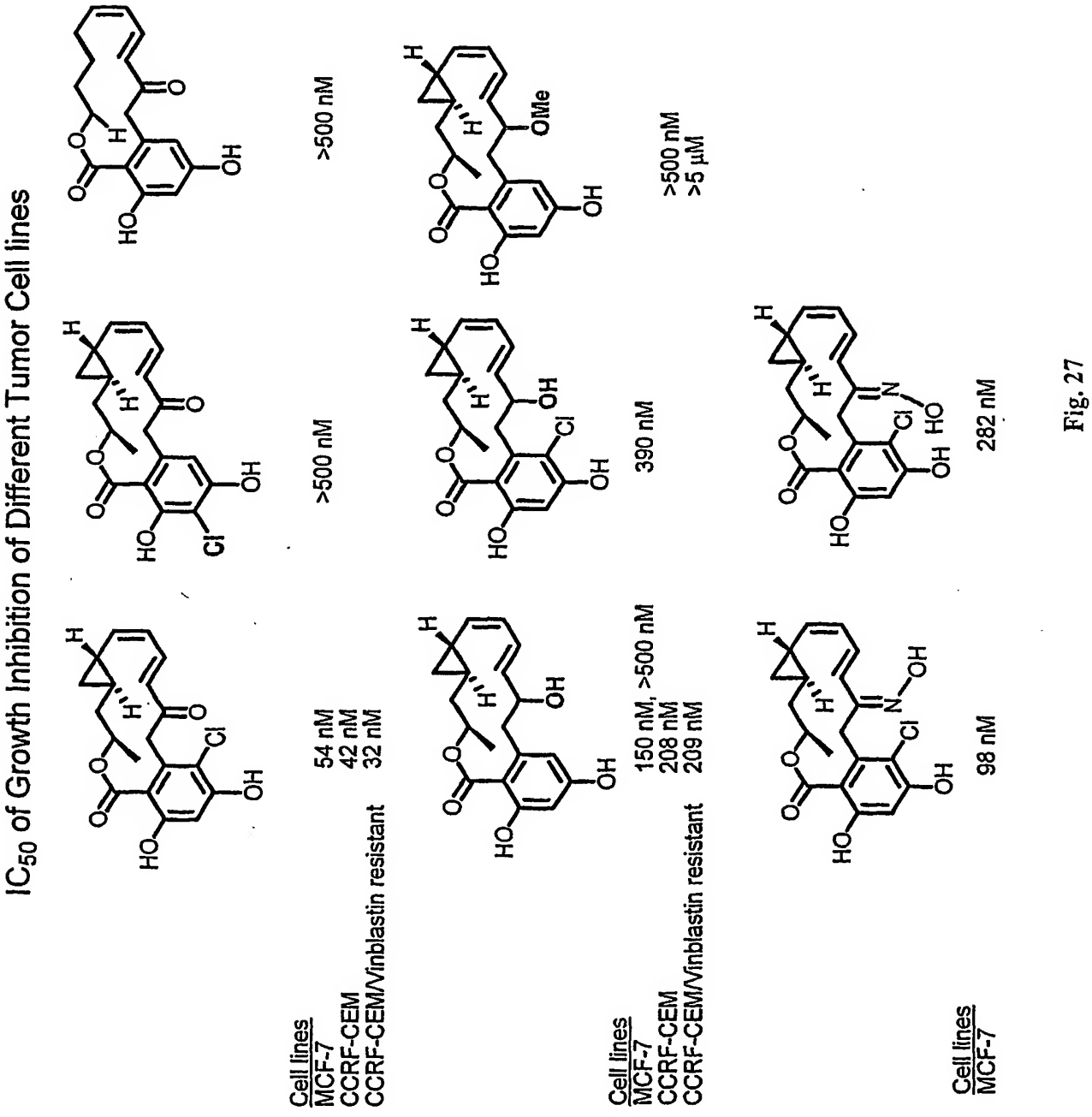

IC50 of Growth Inhibition of Different Tumor Cell lines
Cell lines
MCF-7
CCRF-CEM
CCRF-CEM/Vinblastin resistant
54 nM
42 nM
32 nM
>500 nM
>500 nM
>500 nM
>5 μM
150 nM, >500 nM
208 nM
209 nM
390 nM
98 nM
282 nM

Fig. 27

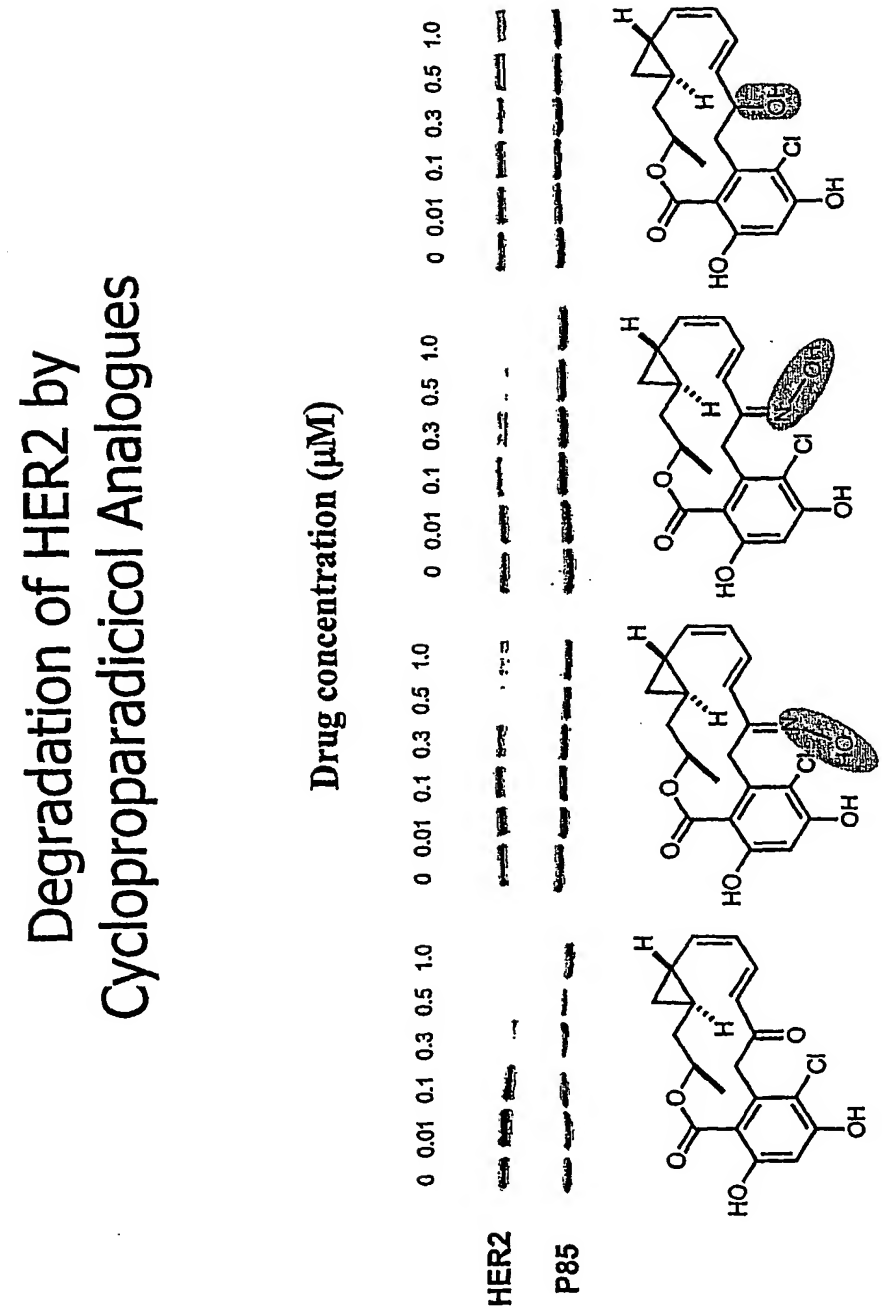

Degradation of HER2 by Cycloproparadicicol Analogues
Drug concentration (μM)
0 0.01 0.1 0.3 0.5 1.0
0 0.01 0.1 0.3 0.5 1.0
0 0.01 0.1 0.3 0.5 1.0
0 0.01 0.1 0.3 0.5 1.0
HER2
P85

Fig. 28

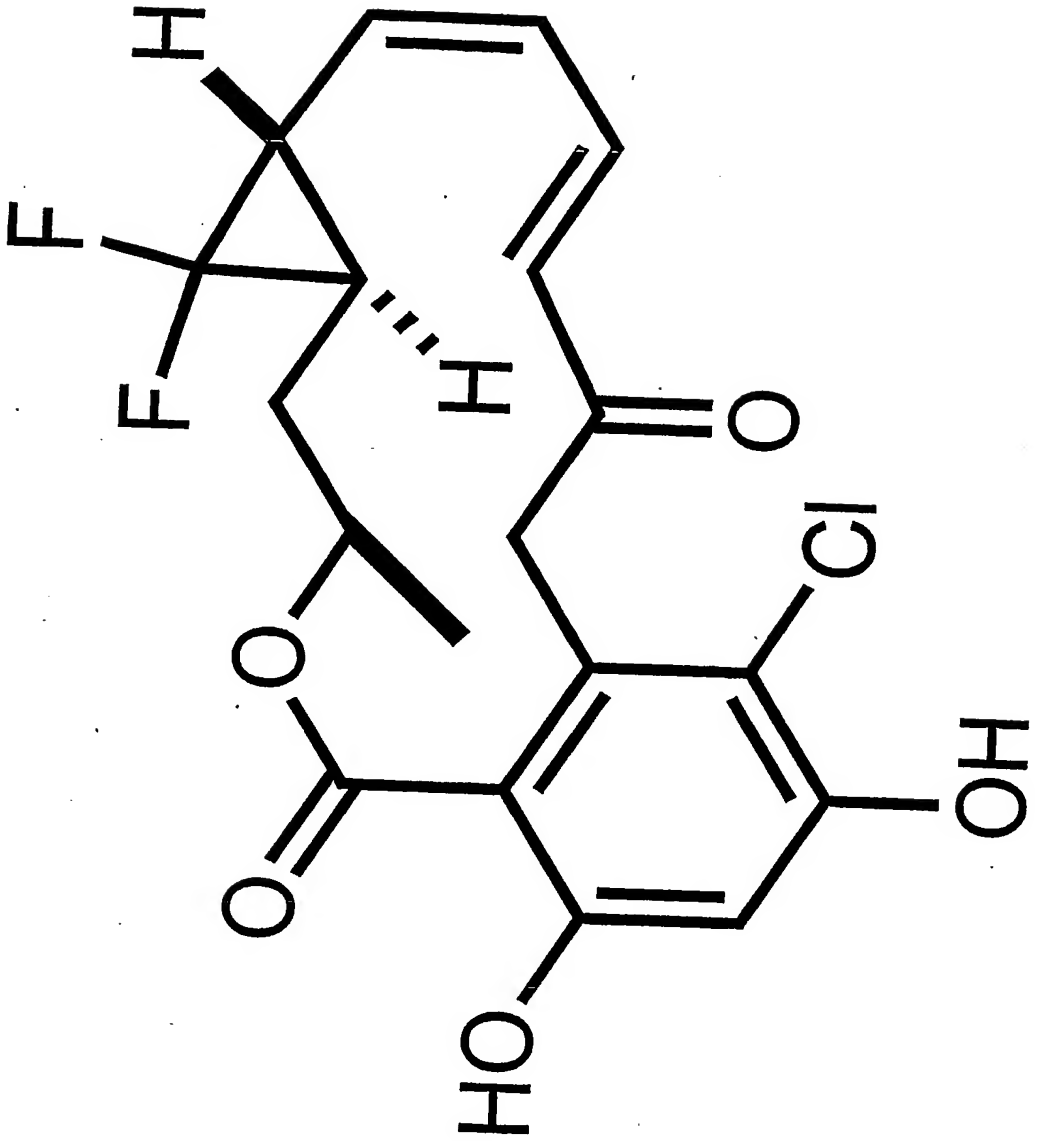


Fig. 29

Cytotoxic effect on CCRF-CEM cell growth by radicicol analogs[a].

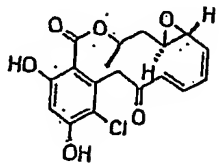
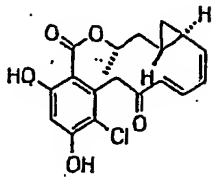
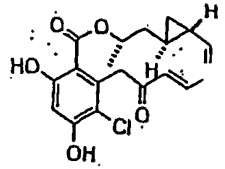
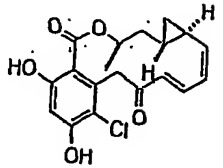
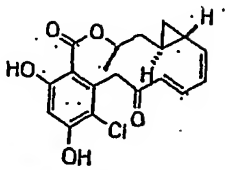
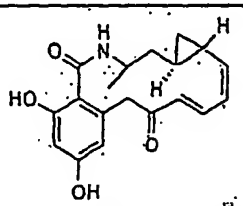
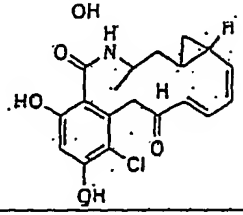
Compound	Structure	Cell growth inhibition (IC_{50} in μM)[b]		
		CCRF-CEM	CCRF-CEM/VBL[c]	CCRF-CEM/taxol[c]
Radicicol (Sigma)		0.055±0.03	0.099 [1.8x]	0.070 [1.3x]
Cyclopropyl 1		4.81	9.84 [2.0x]	7.74 [1.6x]
Cyclopropyl 2		2.34	4.89 [2.1x]	2.89 [1.2x]
Cyclopropyl 3		0.58±0.13	0.87 [1.5x]	0.53 [0.9x]
Cyclopropyl 4 (Cycloproparadicicol)		0.055±0.04	0.041 [0.75x]	0.070 [1.3x]

Fig. 30A

Cytotoxic effect on CCRF-CEM cell growth by radicicol analogs[a]. (Cont'd)

Compound	Structure	Cell growth inhibition (IC_{50} in μM)[b]		
		CCRF-CEM	CCRF-CEM/VBL[c]	CCRF-CEM/taxol[c]
DechloroCyclopropa-radicicol Lactam		>10	>10	ND
Cyclopropradicicol Lactam		>5	>5	ND

[a] Compounds of radicicol and cyclopropradicicol stereoisomers.

[b] Cell growth inhibition was measured by XTT tetrazonium assay after 72-hour incubation for cell growth. (Chou et al., Proc. Natl. Acad. Sci. USA 95: 15798-15802, 1998). Five to eight concentrations for each drug were used. IC_{50} values were determined from dose-effect curves by using a computer program CalcuSyn for Windows by Chou and Hayball (Biosoft, Cambridge, UK, 1997).

c CCRF-CEM/VBL and CCRF-CEM/taxol are the CCRF-CEM sublines that are 320-fold and 42-fold resistant to vinblastine and taxol, respectively. Number in brackets is the fold of resistance of each drug when comparing the IC_{50} values with those of the parent cell line, CCRF-CEM. The results showed that radicicol and cyclopropradicicol stereoisomers are not cross-resistant to vinblastine (typical MDR-Pgp substrate) nor to Taxol.

Fig. 30B

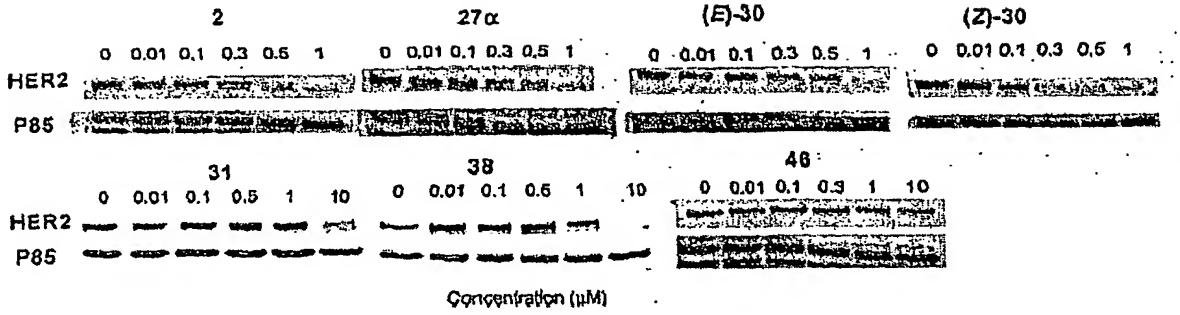


Figure 31: Her2 Degradation Assay